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SER. II, VOL. 16 *December, 1953* NO. 2

# TRANSACTIONS
## OF
# THE NEW YORK ACADEMY OF SCIENCES

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TRANSACTIONS of The New York Academy of Sciences, Series II, Volume 16, No. 2, December, 1953

This publication is distributed to members and is published monthly from November to June, inclusive, by The New York Academy of Sciences, 2 East Sixty-third Street, New York 21, N.Y.

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## *SECTION OF BIOLOGY**

### CELL POPULATION STUDIES ON MOUSE ASCITES TUMORS †

*By* THEODORE S. HAUSCHKA ‡

Cellular variability has presented one of the foremost problems in cancer research. Not only does the origin of a neoplasm involve a cellular change, but radiotherapy and chemotherapy are frustrated by an intangible reserve of metabolic variants. The genetic nature of these changes and their relation to the chromosome constitution of the cancer cell offers a field that can be attacked most profitably through population analysis of ascites tumors.

In contrast with a true panmictic population, where genetic equilibria are maintained through random mating, the gene pool in a malignant cell community is under the haphazard control of selective pressures exerted by the host against competing cell lineages. Population genetics in the Mendelian sense cannot, of course, be investigated in the non-mating clonal descents which constitute a growing neoplasm.

It is, however, possible to elaborate at least some iso-antigenic fluctuations in exact genetic terms, since the inbred host-strain, from which the cancer cells have descended and with which they agree in genotype, can by hybridized with a graft-resistant pure line differing from the malignant graft in one or more blood-group factors or histocompatibility alleles. Methods for the determination of iso-antigenic differences have, by now, become classical through the transplantation experiments of Little [1] and Strong, [2] the serological studies of Gorer, [3] and the linkage tests of Snell. [4, 5]

Our own primary objective has been correlation between immunogenetic and cytologic properties of transplantable mouse cancers. [6, 7, 8, 9, 10] The work to be reported here is based to a large extent on Klein's histologic spectrum of ascites tumors [11, 12, 13] including strain-specific, as well as genetically indifferent lymphomas, sarcomas, and carcinomas (TABLE 1) and special sublines which have been derived from these oncotypes.

In our analysis of neoplastic cell-populations we have been concerned with:

(1) chromosome number in relation to histocompatibility;

(2) experimentally induced changes in antigenic properties;

(3) distinct clonal components of tumors bearing on the question of stem-cells and their function as the principal progenitors of growing cancers.

These problems have been approached through mutually supportive cytological, immunogenetic, and microisolation techniques.

#### *Inverse Relationship Between Chromosome Ploidy and Host Specificity*

All tumors investigated in collaboration with Levan for chromosome number distribution [6, 10, 14] were found to be complex chromosomal mosaics with modal values at or near diploid (= 40 in the mouse) or in the tetraploid region between 70 and 90. These findings are in quantitative agreement with the values of DNA/cell determined at other laboratories. [12, 15] Variations in chromosome number from 40 to 148 are pictured in PLATE 1; A–H. The mitotic irregularities responsible for these wide fluctuations are endoreduplication (PLATE 1, I), multipolarity, lagging, non-disjunction, and bridging (PLATE 1, J).

* The Section of Geology and Mineralogy held a meeting on November 2, 1953 at which a paper, entitled "The Carbon 14 Method of Age Determination," was read by J. Laurence Kulp. No abstract of this paper had been received at the time of publication.

† This paper, illustrated with lantern slides, was the first of two papers presented at the meeting of the Section on November 9, 1953. The work was supported, in part, by grants from the National Cancer Institute, the Damon Runyon Memorial Foundation, and the American Cancer Society.

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TABLE 1

CHROMOSOME NUMBER AND HOST SPECIFICITY IN MOUSE ASCITES TUMORS *

| Chromosome Number Mode | Carcinomas | Sarcomas | Lymphomas | Strain Specificity |
|---|---|---|---|---|
| 40-42 | TA3 (Hauschka) | | A#2<br>DBA<br>6C3HED | Specific for strain of origin |
| 45-47 | Ehrlich (Lettré) | Landschütz I, II | | Non-specific |
| 70-76 | Krebs 2 stock (1953)<br>K2 sublines A, B, C<br>K2 clones D, E<br>Ehrlich stock (1953)<br>Ehrlich clones 2, 4, 11<br>TA3 (Lederle) | MC1M<br>S37 | | Non-specific |
| 80± | 15091a<br>Krebs 2 stock (1951)<br>Ehrlich stock (1951) | MC1A | A#1 | Non-specific |
| 86-92 | Ehrlich clone 1<br>EL88 (Kaziwara) †<br>EL89<br>EL90 | | | Non-specific |

* Source references and other details concerning most of these tumors have been listed by Klein[12] and by Dunham and Stewart.[26]

† EL88, EL89, EL90 represent hypertetraploid derivatives from Lettré's Ehrlich, which arose at our laboratory through endoreduplication.

The chromosome number fluctuations are expressive of considerable inequalities within each neoplastic cell-population. Although each tumor has its own individualistic chromosome idiogram and one cannot discern any cytologic anomalies specific for cancer, there are (in tune with Boveri's speculations[16]) two over-all differences in distribution pattern between cancer and normal tissue:

(1) Aneuploid and heteroploid nuclei are much more frequent in neoplasms[6, 10, 14, 17, 18, 19, 20] than in normal adult or embryonic material,[14, 21, 22] or in tissue cultures of normal mammalian cells,[23] where the diploid number always predominates. Timonen's[24] extensive sub-diploid counts in normal human endometrium could not be confirmed by Sachs[22] in identical tissues from man, rat, and vole.

(2) Most transplantable tumors are characterized by aneuploid chromosome number modes, suggesting that such neoplasms may derive some of their growth advantages from genic imbalance.

An apparently constant correlation has been established between modal chromosome ploidy and host-specificity.[7, 9] All of the near-diploid tumors examined consistently regressed in mice of foreign strains, while the ascites in the heteroploid category invariably gave lethal takes in from one to all seven foreign genotypes tested. This trend appeared to be independent of the tumor's histologic and genetic origin, transplantation history, virulence and method of induction.

The generalization, that tetraploid and aneuploid growths are more or less compatible across genetic barriers while near-diploid tumors are strain-specific (TABLE 1), has recently been subjected to a more critical test with three cytologically distinct lines of TA3. This mammary adenocarcinoma originated spontaneously in one of our A/Ha♀♀ in 1949, was carried as a

solid tumor for 34 transplant generations in A-mice, was converted into an ascites in June 1951, and has passed through 151 serial transfers in the A-strain. The shift toward hyper-diploid occurred about 30 passages ago. The change to hypo-tetraploid was noted in TA3 ascites material received from Doctor Koprowski via Doctor Klein who maintains another heteroploid branch of TA3. The inter-strain transplantation results for these three variants of TA3 are here presented for the first time (TABLE 2); they demonstrate rather conclusively that indeed there exists an inverse relationship between chromosome ploidy and histocompatibility. This fact does not detract in the least from the validity of classical transplantation genetics. On the contrary, it provides plausible chromosomal mechanisms for two phenomena which have puzzled geneticists:

(1) During serial passage, a tumor may lose not just one, but several antigens as a bloc – thereby finally becoming non-specific.[25]

(2) Changes in the opposite direction, that is to say increases in tissue specificity, have been very rare.[1]

These two observations are in accord with the periodic chromosome number changes in some of the ascites tumors. The shifts have proceeded either slowly or suddenly from diploid to heteroploid, never in the opposite direction.

A simple duplication of the diploid chromosome set is certainly *not* the essential factor in the loss of antigenicity. If this were the responsible mechanism, the rare tetraploid cells present in all diploid populations would be selected out after inoculation into refractory hosts; but this does not happen.[9] On the other hand, several tumors with aneuploid modes only a little above the normal diploid 40 exhibited a wide host range (TABLE 1).

The reduced antigenicity of these aneuploid as well as the near-tetraploid tumors is interpreted as resulting from changes in genic equilibrium. In a heteroploid cell, more than two loci may be available for some antigenic genes. The antigenic alleles can, therefore, function in dosages other than those to which they are restricted in a diploid nucleus with consequent alteration or even extinction of their specific end-products. Loss of specificity may thus be viewed as immunologic selection of the least antigenic types from among the viable chromosomal variants in a neoplastic population.

### *Increased Antigenic Specificity of Tumors*

The number of histocompatibility genes (H-factors) governing antigenic constitution, and thereby transplantability, has been worked out for several of our strain-specific ascites tumors (TA3, A♯1, A♯2, DBA, 6C3HED, MC1M) in relation to certain foreign mouse-stocks. These tests have established convenient base-lines for later recognition of antigenic changes in either the tumors or the hosts, and for possible correlation with concomitant shifts in the chromosome constitution of the tumors. Pursuing the question of possible immunogenetic differences between various implantation sites in their response to one

TABLE 2

CHROMOSOME PLOIDY AND HOST-SPECIFICITY IN THREE SUBLINES OF TA3 ASCITES

| Subline of TA3 adenocarcinoma (A-strain origin) | Percent diploid cells | Percent aneuploid cells | Percent tetraploid cells | Per cent lethal takes in various strains | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | A | C3H | C58 | DBA | Y/He | C57 | Swiss | 129 |
| Diploid TA3 | 46 | 48 | 6 | 100 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| Hyperdiploid TA3 | 6 | 93 | 1 | 100 | 74 | 31 | 12 | 25 | 31 | 94 | 25 |
| Hypo-tetraploid TA3 | 2 | 94 | 4 | 100 | 100 | 100 | 94 | 83 | 71 | 70 | 11 |

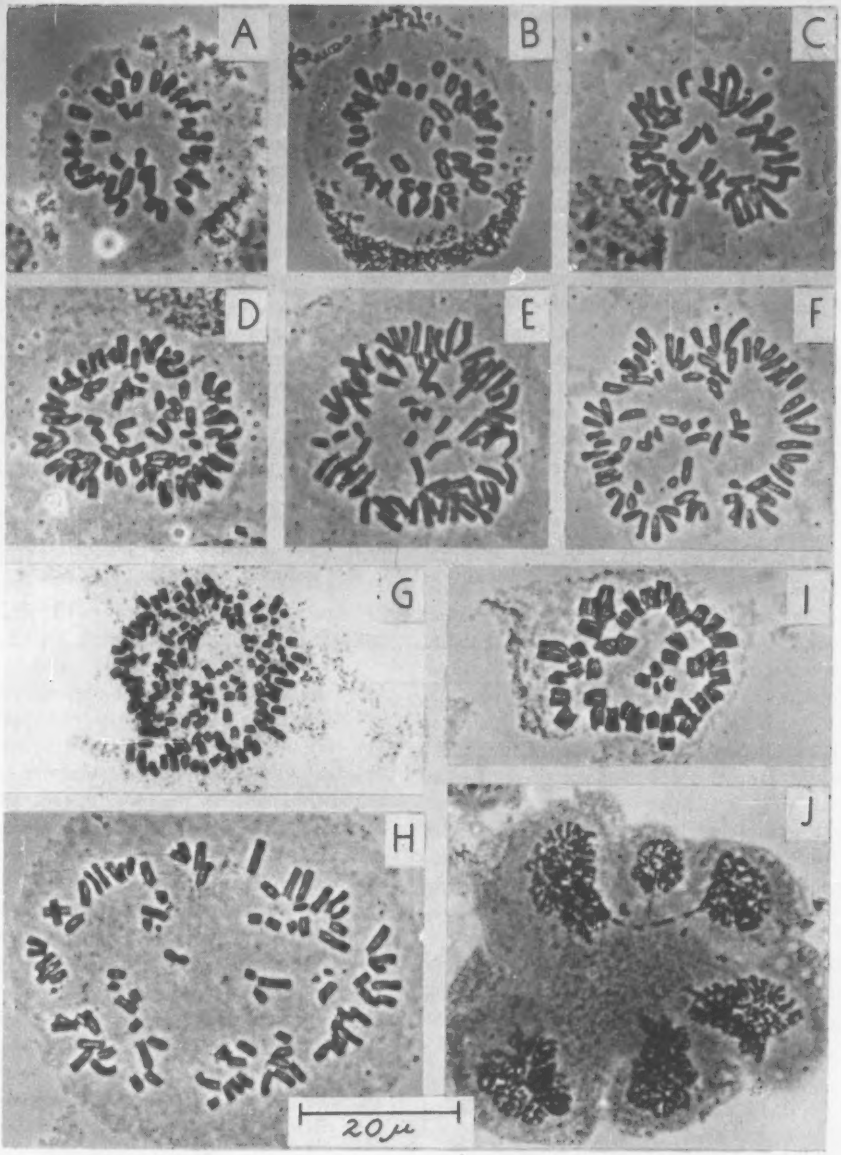


PLATE 1

**Variations of chromosome number in mouse ascites tumors. (Phase contrast photos of cells fixed with aceto-orcein and pressed out on siliconed slides.) A—E. Metaphases of the TA3 mammary adenocarcinoma with 40, 41, 45, 82, and 75 chromosomes. F and H. Ehrlich clone 1, metaphases with 83 and 90 chromosomes. G. Hypo-octoploid cell from Ehrlich clone 2 showing exact duplication of the clone's modal chromosome number of 74. I. Endo-reduplication in a cell of the hyperdiploid Lettré Ehrlich showing 45 four-stranded chromosomes. J. Hexapolar telophase in the near-tetraploid Krebs-2 ascites. Six unequal daughter nuclei, a persisting bridge, two laggards, and abortive cytoplasmic cleavage are evident.**

and the same tumor, we found that the DBA lymphoma required 2 H-factors intraperitoneally, and 3 or 4 H-factors subcutaneously in (DBA × C3H) BC hybrids.[27] On the other hand, growth of two other lymphomas (6C3HED and A#1) fitted a 1-gene ratio in both the intraperitoneal and subcutaneous sites in (C3H × DBA) and (A × C3H) backcross and $F_2$ genotypes (unpublished data of Hauschka, Kvedar, and Grinnell).

It has been demonstrated by Strong[25] and emphasized by Little[1] that spontaneous alterations in the immunogenetic properties of tumors almost always proceed in one direction, *i.e.* toward reduced specificity, through loss of one or several H-factors. Barrett and Deringer[28, 29, 30] have developed an experimental procedure whereby antigenic changes may be regularly induced; but the direction or magnitude of these enforced modifications is unpredictable. All that is required is to pass a strain-specific tumor, which has been propagated in the strain of origin, temporarily through $F_1$ hybrids between the inbred home-stock and a foreign pure line. The Barrett's backcross tests with their C3HBA carcinoma showed decreased specificity after $F_1$ passage in comparison with BC take percentages after uninterrupted home-strain passage. Our similar experiments with the TA3 adenocarcinoma and the DBA lymphoma have given results with an opposite trend. The modified percentages of lethal backcross takes obtained by us usually fitted segregation ratios for increased gene-requirements.[27]

It is probable that immunologic selection, rather than antigenic adaptation, accounts for the induced genotypic conditioning of

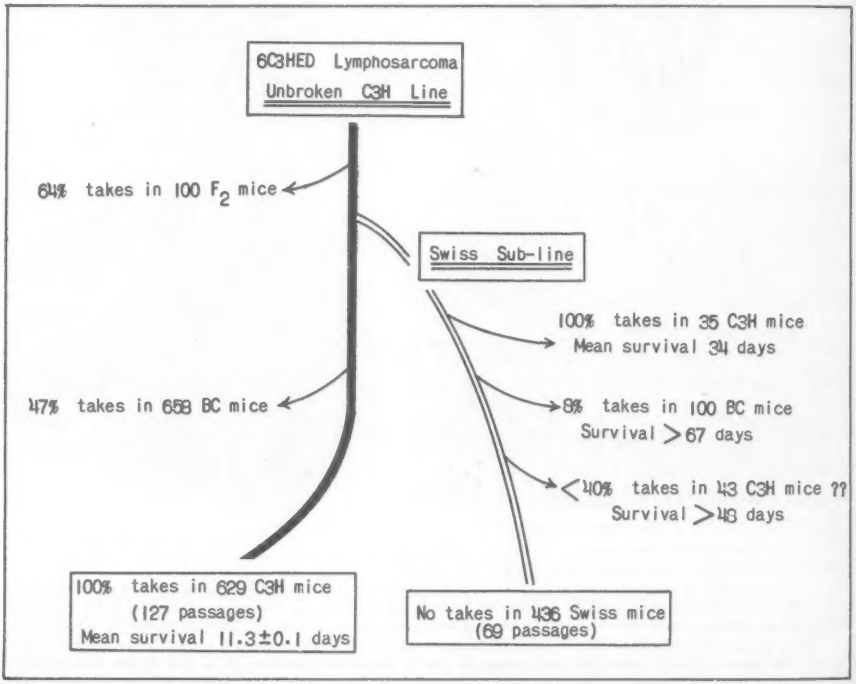


FIGURE 1. Increased antigencity (?) of the 6C3HED lymphosarcoma ascites acquired during serial passage in a foreign resistant strain.

grafts during $F_1$ residence; for the change in transplantation potential is irreversible, becomes fully manifest after a single $F_1$ passage, and exhibits rather close agreement between the new take-percentages and Mendelian expectation.

A possibly analogous case of increased specificity is currently developing in one of our sublines of the 6C3HED lymphosarcoma (FIGURE 1). Instead of passing this C3H tumor through $F_1$ mice where it would be 100 per cent lethal, it was nursed along for 70 weekly passages in the foreign Swiss strain, where it has always regressed after good temporary growth. The main line of this virulent neoplasm, estrogen-induced in a C3H mouse by Gardner,[31] has been carried by us in the ascites form since it was received from Doctor Klein in May 1951. During 127 weekly serial passages in C3H/St it has killed all hosts; average survival time was 11.3 ± 0.1 days (standard dev. 1.8). Histocompatibility tests for C3H passages 34, 35 and 90-112 gave a 1- or 2-gene relationship with A-strain and an excellent fit (47.3 ± 1.9 per cent) for a 1-gene difference in (C3H × DBA) BC hybrids.

The Swiss subline of the tumor was branched off from our 56th main line passage in July 1952. Ascites from the 18th–23rd Swiss transfers re-inoculated into C3H/St mice was still lethal, but mean survival time was significantly increased to 33.9 ± 1.3 days (standard dev. 6.4). Injection of 150 million lymphosarcoma cells from the 64th–67th Swiss passages into each of 100 (C3H × DBA) BC hybrids has given only 8 takes, instead of the expected 50 deaths, and all of these BC mice are still living 70 days after inoculation in a current experiment.

This apparent increase in antigenicity applies not only to resistant backcross mice, but also to the C3H home-strain which is becoming more and more refractory to its own lymphoma from the Swiss subline. The partial loss of tissue-affinity for C3H might be ascribed to cumulative minor antigenic changes in the home-stock, with which the cell population in continuous C3H passage was able to keep pace, while the Swiss subline was making adjustments to immunologic and/or metabolic selection of a far more drastic order. H-factor mutations in the C3H hosts were, however, ruled out by lethal takes in C3H mice of frozen 6C3HED ascites from one of the early passages after 17 months storage in our tissue bank at −40° C.

TABLE 3

TAKES OBTAINED WITH LOW CELL-DOSAGES OF ASCITES TUMORS

| Tumor | Host strain | Mean inoculum | Number of mice | Number of takes |
|---|---|---|---|---|
| Ehrlich carc. | Swiss | 27 cells | 21 | 13 |
| Krebs 2 carc. | Swiss | 14 " | 20 | 6 |
| S-37 sarc. | Swiss | 14 " | 21 | 0 |
| TA3 carc. | A/Ha | 30 " | 20 | 19 |
| A#1 lymph. | A/Ha | 22 " | 20 | 0 |
| A#2 lymph. | A/Ha | 14 " | 21 | 0 |
| DBA lymph. | DBA/2 | 22 " | 20 | 16 |
| DBA lymph. | DBA/2 | 3 " | 21 | 15 |
| 6C3HED/Ha | C3H/St | 15 " | 20 | 19 |
| 6C3HED/KL | C3H/St | 15 " | 20 | 1 |
| MCIM sarc. | $C3H_f$/He | 24 " | 20 | 0 |

Morphologically, the attenuated 6C3HED cells from Swiss donors are identical with those of the virulent main line. The distribution of chromosome ploidy and the characteristic nuclear lobation are still approximately the same as described for the main line a year ago.[32]

The two branches of the tumor provide a promising dual test-system for experimental decision between alternative hypothetical mechanisms responsible for the antigenic (?) transformation.

### *Tumor Clones Derived from Single Transplanted Cells*

Prior to the first successful transplantation of single malignant leukocytes by Furth and Kahn,[33] it was generally believed that several thousand cells were necessary to transmit the then available mouse carcinomas, sarcomas, and leukemias. Ishibashi[34] has recently perfected a method for

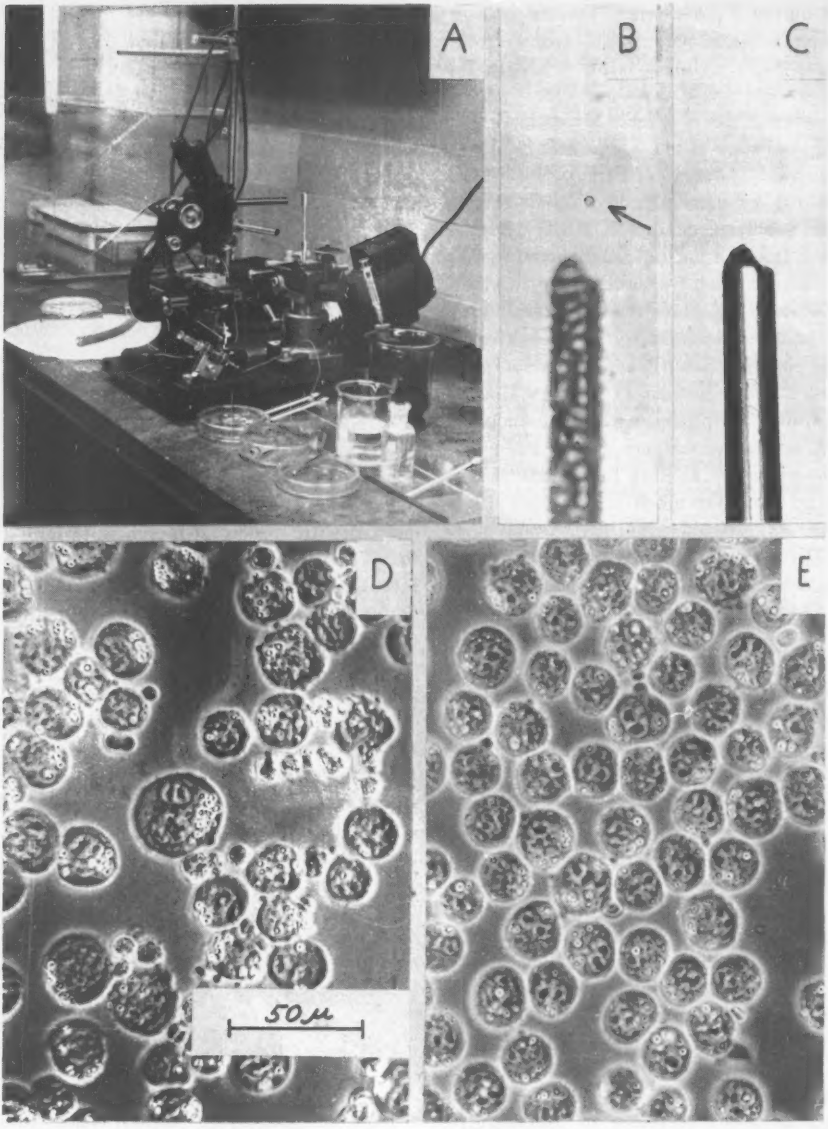


PLATE 2

Isolation of single cancer cells and establishment of clones. A. Apparatus used for isolating and injecting single cells. B and C. Cell (arrow) in hanging drop and micropipette out of focus in moist-chamber. Tip of same pipette after cell and surrounding liquid have been picked up. D. Heterogenous cell-population of the Ehrlich stock tumor. E. Uniform clone derived from a single cell of the former. (D and E are phase-contrast photos of living cells.)

the one-cell inoculation of the Yoshida rat sarcoma. The primary object of these earlier isolation experiments was to show that at least one living, intact cell was essential for transmitting the mammalian tumors in question, and that neither cell-free material nor experimentally damaged cells could initiate neoplasias of corresponding type in a mouse or rat.

The rat tumors descended from single cells were, according to Yoshida,[20] exactly the same as the original ascites propagated with standard injections of 50 million cells, except that they required about ten days longer for their appearance. In the absence of clear cytologic differences between their clones and stock tumors, Yoshida[20] and Sato[35] have questioned the existence of stem-cell lineages[19] as progenitors of the neoplastic population.

The wide range of chromosome numbers observed by us in most ascites populations,[6, 10] the spontaneous transitions of individual tumors from near-diploid to polyploid which we witnessed four times (TA3, EL88, EL89, EL90) during the past year, and the inverse relationship between the degree of heteroploidy and histocompatibility,[9] suggested a re-examination of the stem-cell hypothesis through comparison of single-cell clones. In preparation for this work, low cell-dosages of 10 different ascites tumors, ranging from 3 to 30 cells per mouse, were inoculated into animals of appropriate genotype which always succumb to the standard inocula of around 20 million cells. The results of these dilution tests (TABLE 3) were positive in 7 out of 11 separate trials. For the critical single-cell isolations,[36] the Krebs 2 and Ehrlich carcinomas were chosen because of their large cell-size and cytologic variability, and 6C3HED lymphosarcoma because of the high take-percentage obtained with only 15 cells per mouse. The Japanese technique[34] was simplified for our purposes as follows: The ascites was diluted 1:100,000 to 1:500,000 in a medium consisting of 32 parts special mouse-Ringer (8.1 g. NaCl, 0.2 g. KCl, 0.2 g. $CaCl_2 \cdot 2H_2O$ per liter of triple-distilled water, buffered to pH 7 with 0.04 M sodium phosphate buffer) to 1 part of centrifugal supernate prepared from the ascitic fluid of the donor mouse. At this great dilution, more than half of the microdroplets suspended above a moist-chamber contained no cells at all. Hanging drops with only one cell were picked up with a dry capillary pipette controlled by means of an Emerson micromanipulator at 200 × magnification (PLATE 2, A–C). The cell was then delivered intraperitoneally into an infant or adult mouse by perforating the body-wall with the glass micropipette and applying air-pressure from a greased 2 cc. syringe. No takes have as yet been obtained with lymphosarcoma 6C3HED; but the positive results with the Krebs 2 and the Ehrlich carcinomas have been repeated several times and are nearly identical from test to test. Nineteen tumor clones have been recovered from 212 mice within 16 to

TABLE 4

PER CENT DISTRIBUTION OF CHROMOSOME NUMBERS IN KREBS 2, EHRLICH, AND CLONAL LINES

| Tumor or Clone | Chromosome Number | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 34-69 | 70 | 71 | 72 | 73 | 74 | 75 | 76 | 77 | 78 | 79 | 80 | 81 | 82 | 83 | 84 | 85-100 | > 100 |
| K2 stock | 15 | 1 | – | 1 | 3 | 1 | 3 | 4 | 7 | 6 | 4 | (9) | 8 | 8 | 3 | 4 | 12 | 11 |
| Clone K2D | – | 2 | 6 | 10 | 10 | 4 | (22) | 4 | 8 | 4 | 2 | 6 | – | 2 | – | – | 2 | 18 |
| Clone K2H | – | 12 | 4 | 6 | 10 | (18) | 14 | 14 | 2 | 2 | – | – | – | – | – | – | 2 | 16 |
| Ehrlich stock | 4 | – | – | 2 | 2 | – | 2 | 4 | 4 | 4 | 8 | (18) | 8 | 16 | 10 | 2 | 14 | 2 |
| Clone E1 | – | – | – | – | – | 2 | – | – | – | – | – | 20 | 4 | 12 | 6 | 16 | (40) | – |
| Clone E2 | 2 | 2 | 2 | – | 6 | 16 | (22) | 10 | 8 | 4 | – | – | – | 2 | 4 | – | 2 | 20 |

25 days after injection of a single cell from the Ehrlich or Krebs 2 tumors. The increase in cell-number during this period averaged 270 million, *i.e.*, about $2^{28}$, or a mean of one mitosis per cell every 17 hours. Three-day-old infant Swiss mice of both sexes gave 15 per cent takes, and were approximately 7 times more susceptible than young adults.

The clones were cytologically more uniform and usually more virulent than the stock tumors from which they were isolated. A comparison between the stock Ehrlich (PLATE 2, D) and Ehrlich clone 7 (PLATE 2, E) reveals impressive homogeneity of the latter. The increased virulence could result from the larger proportion of cells with continued mitotic capacity and also from immunologic selection, since the chance for a successful take is best for those cell-types which are least responsive to the host's defense reactions.

Each clone is characterized by a stem-line ideogram which may be numerically, structurally, and functionally distinct from the chromosome constitutions in other clones (TABLE 4). The chromosome-numbers of the individual cell-lineages cannot remain rigidly fixed because of mitotic irregularities; but the numerical ranges are definitely narrowed; and distinct new modes may become established. It is perhaps significant that the most frequent chromosome-number modes were sub-tetraploid in the neighborhood of 75, and that both the stock Ehrlich and stock Krebs 2 have gradually shifted from their former tetraploid modes toward this aneuploid number. Only one out of ten Ehrlich clones (E1 in TABLE 4) was decidedly hyper-tetraploid, and has persisted as such for over 50 serial transfers during a period of eight months.

The concept of stem-cells from which each tumor propagates itself appears to have been strengthened through the isolation experiments. This is borne out by the observed differences between stock-tumors and clones, the divergencies between the cell-lineages themselves, and the preferential recovery of certain aneuploid chromosome-numbers in the mid-70's and near 90, which appear to represent the most viable portion of the population curve. Most of the isolated cells are nonviable, even under the favorable conditions of transfer into newborn mice. Although, by morphologic criteria, they are cancer cells they do not have the stem-cell's capacity of unlimited self-reproduction and are, therefore, functionally nonmalignant.

### *Conclusions*

1. Fluctuation in chromosome-number, equivalent to genetic variability, occurs over a much wider range in mouse cancers than in normal tissues of the mouse, where the diploid number of 40 always predominates. Most transplantable tumors are characterized by aneuploid chromosome-number modes.

2. Heteroploid growths are more or less compatible across genetic barriers, while diploid and near-diploid tumors are host-specific. This generalization, based on our earlier comparison of 16 unrelated transplantable mouse and rat neoplasms, has now been consolidated by tests with three sublines of the TA3 mammary adenocarcinoma differing from one another in chromosome ploidy.

3. The antigenicity of some neoplasms may be increased experimentally by temporary passage through hybrids between the mouse-stock of origin and a resistant strain. In lymphosarcoma 6C3HED the isoantigenic structure appears to have been altered to the point of greatly reduced virulence for the genotype of origin by prolonged serial transfer through foreign resistant hosts.

4. Only about 15 per cent of cancer cells can be successfully isolated and transplanted from favorable tumors; they give rise to pure clones more uniform and often more virulent than the original stock-

tumors, while the remaining cells fail to take. The soundness of the stem-cell concept, as well as the relative frequency of nonmalignant cancer cells, is thereby indicated.

### *Acknowledgments*

Barbara J. Kvedar, Sarah T. Grinnell, Elizabeth G. Brown, and David A. Hungerford have skillfully participated in various technical phases of our program. I am indebted to Doctor G. Klein for information concerning his subline of TA3, to Doctor A. Levan for his unpublished data on the Landschütz tumors, and to Doctor J. Schultz for stimulating discussions. Doctor H. Lettré has kindly sent us the hyperdiploid line of the Ehrlich ascites.

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## DYNAMICS OF RESPIRATORY PIGMENTS OF ASCITES TUMOR CELLS*

*By* BRITTON CHANCE †

The ascites tumor cells represent, at present, the most convenient sample of intact mammalian tissue that can be used for spectrophotometric studies of the dynamics of intracellular enzyme reactions. For such studies, our methods require a free cell suspension with which the substrates and inhibitors may be rapidly mixed and into which these substances will rapidly penetrate. We can obtain such free tumor cell suspensions in adequate quantities from a few mice for our spectroscopic studies. The recent developments made by Doctor Hauschka in the growth of these tumor cells from single clones provides us with a material that is nearly homogeneous morphologically and is well characterized and controlled genetically. A further requirement for our studies is that the cells be rugged enough to withstand the turbulence caused by rapid flow apparatuses, and in addition, they must be spherical in shape in order that orientation of the flow pattern should not affect the light transmission.

Working with less well characterized materials a few years ago we published a preliminary note on spectrophotometric studies of respiratory pigments in suspensions of these tumor cells.[1] We have now been able to extend and to refine considerably the spectroscopic data on the respiratory pigments of these tumor cells. We have verified our previous result concerning the relative abundances of cytochrome c and cytochrome oxidase in these cells, and show cytochrome c to be present to as great or greater extent than it is present in tissues known to contain a large amount of cytochrome c, for example, mammalian muscle. Other pigments in addition to the cytochromes have been found to be present in these tumor cells in normal amounts, for example, flavoprotein and reduced pyridine nucleotides.

Direct experimental evidence for the potentially high level of activity of the oxidative enzyme systems of these tumor cells is provided by a rapid kinetic study of the oxidation of reduced respiratory enzymes upon admitting in oxygen. This is accomplished by means of a special rapid flow apparatus in which anaerobic cells are mixed in a few milliseconds with a small amount of dissolved oxygen. Spectrophotometrically, we can measure the very rapid oxidation of the cytochromes and flavoprotein indicating the great potentialities of this oxidase system. On the other hand, one observes a very slow rate of oxygen uptake in steady state respiration and this would presumably be attributed to a lack of dehydrogenase activity.

Our attention has, therefore, been turned to the kinetics of the reduced pyridine nucleotides in the intact cell which we can measure directly with our sensitive spectrophotometers. In the ascites tumor cells one finds a large proportion of the absorption due to reduced pyridine nucleotides to have a maximum at 325 to 330 millimicrons in contrast to the position of 340 millimicrons observed for reduced pyridine nucleotides in solution. This is tentatively attributed to a binding of the reduced pyridine nucleotides to a protein. Such a shift in absorption spectrum of reduced pyridine nucleotides has been observed *in vitro* in the presence of the enzyme lactic dehydrogenase, an enzyme that is present in these tumor cells.[2] Our spectroscopic data interpreted in this manner lead us to the conclusion that the larger part of the reduced pyridine nucleotides are bound to an enzyme such as lactic dehydrogenase in the tumor cells.

* This paper, illustrated with lantern slides, was the second of two papers presented at the meeting of the Section on November 9, 1953.

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In the oxidized cell a distinctive absorption band having a maximum of 365 millimicrons is obtained upon adding iodoacetate. A very similar absorption band is obtained *in vitro* when iodoacetate is added to the DPN compound of the glyceraldehyde 3-phosphate dehydrogenase.[3] We would, therefore, conclude that this compound exists in the oxygenated tumor cell also.

A rather interesting effect observed with these tumor cells is the lack of stimulation of respiration upon adding substrates, in fact it has already been observed that glucose can inhibit the respiration of these cells.[4] In our spectroscopic studies the addition of glucose to the aerobic cells causes reduction of an appreciable amount of pyridine nucleotide. Interestingly enough this reduction of pyridine nucleotides causes no sustained increase of respiration. Making the assumption that pyridine nucleotides reduced by glucose are accessible to the respiratory system, we then conclude that the rate-limiting step in the tumor cell respiration is not due to a deficiency of reduced pyridine nucleotides as a consequence of low dehydrogenase activity, but the weak link may lie between the reduced pyridine nucleotides and the flavoprotein. Our methods do not tell us whether a physical or a chemical factor is involved therein.

In summary, these studies give a clear-cut example of how free tumor cell suspensions may be used in studies of the spectra of their respiratory pigments, the kinetics of their reactions with oxygen and the activity of the dehydrogenase systems that interact with pyridine nucleotides.

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## SECTION OF PHYSICS AND CHEMISTRY

# SOME FRACTIONATION EFFECTS INVOLVING THE ISOTOPES OF HYDROGEN*

By MAXWELL L. EIDINOFF†

The use of deuterium and tritium as tracer atoms for hydrogen has been widely used to explore aspects of chemical reactions of both general and biochemical interest.[18, 26] The extent of isotopic selection in chemical studies can usually be measured by the use of either deuterium or tritium together with light hydrogen. However, in *in vivo* metabolic studies, dilution with light hydrogen during the experiment makes it necessary in many cases to use both deuterium and tritium simultaneously as tracers and to compare the ratio of tritium to deuterium in the products of metabolism with that in the precursor compounds in order to assay the extent of isotopic selection.[9, 30]

The theoretical basis for isotope fractionation effects in equilibrium and reaction rate constants has been extensively reviewed in the literature.[1, 2, 3, 14, 29] Differences in the zero point vibrational energies of bonds involving the isotopic species are, in many cases, the principal factors leading to isotopic selection. In general, equilibrium constant calculations have agreed well with experiment for reactions in which the vibrational frequencies of reagents and products could be properly assigned. The calculation of rate differences for reactions involving protium, deuterium, and tritium requires a precise knowledge of the rate-determining steps and the molecular constants, including the vibrational frequencies for the activated complex in these steps. Unfortunately, our knowledge of the precise nature of these frequencies is usually inadequate to make possible a quantitative calculation.

The order of magnitude of the rate differences in a reaction which involves the rupture of a carbon-hydrogen bond can be estimated if simplifying assumptions are possible. Assume that the differences in activation energy are caused by the differences in zero point vibration energy for the C-H stretching frequency. In this case the smallest activation energy is required for the C-H rupture and largest for the C-T. The order of magnitude of these differences may be seen by the following calculations: Taking the C-H and C-D stretching frequencies as 3060 and 2255 $cm.^{-1}$ respectively, and estimating the C-T vibration as 1890 $cm.^{-1}$, the zero point energy for C-H is lower than that for C-D and C-T by 1150 and 1670 calories per mole respectively. The ratio of velocity constants for isotopic reactions is sometimes written in the approximate form:

$$k_1/k_2 = Ae^{-\Delta E/RT}$$

where $\Delta E$ is the difference in activation energy for the two reactions. If it is now further assumed that A is unity and that the differences in activation energies are equal to the differences in the zero point energies calculated above, then the rate constant in the case of the C-H bond would be greater than that for the C-D and C-T bonds by the factors 6.8 and 16.3 respectively. The rate constant in the case of the C-D bond would thus be 2.4 times greater than that for the C-T bond. These calculations serve to indicate the isotopic fractionation effects due solely to differences in the zero point energy for the reacting molecules, other factors being ignored.

The results of some experiments involv-

* This paper, the first of two papers in a symposium on Fractionation Effects Involving the Isotopes of Hydrogen, was presented at a meeting of the Section on November 10, 1953. The work reported herein was supported by grants-in-aid from the Atomic Energy Commission AT (30-1)—910.

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ing isotopic fractionation effects arising from rate differences will be reviewed. The first group of examples are of general chemical concern; the second group deals with two *in vitro* reactions of biochemical interest; and the third group deals with *in vivo* studies. The literature in the first field, that of reactions of general chemical interest, is now very extensive and no attempt to cover this field adequately will be made.

*I. Reactions of General Chemical Interest.*

*1. Liberation of hydrogen at the cathode during electrolysis.* The enrichment of deuterium in the medium during the extensive electrolysis of aqueous solutions was observed in the initial stages of deuterium research. At smooth platinum electrodes the deuterium content of the hydrogen gas is 6-8 times smaller than that in the liquid medium.[11, 32] At a platinized platinum electrode the observed isotopic concentrations are very close to those corresponding to thermodynamic equilibrium in the exchange reaction between HD, $H_2O$, $H_2$ and HDO. This result is in accord with the accepted view that isotopic exchange is rapid at a platinized surface. Fractionation factors close to 14 were observed for protium relative to tritium in an alkaline medium using smooth platinum electrodes.[6, 7] The equilibrium constant for the gas reaction:

$$HT + H_2O \rightleftarrows H_2 + HTO$$

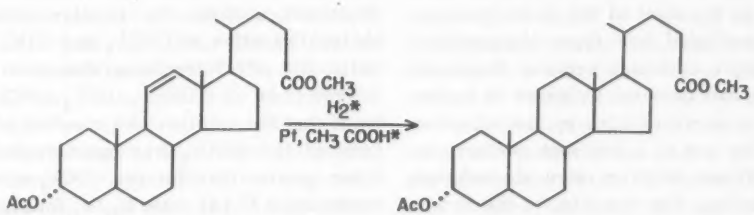
was found to be 6.47 at 20° in good agreement with calculated value of 6.24.[4, 19] Consequently, the protium-tritium ratio for the hydrogen gas in the electrolyses reported above is slightly more than twice as large as that corresponding to thermodynamic equilibrium prevailing at the electrode surface. A similar factor close to two holds for the protium-deuterium ratio. Fractionation factors of 7.2 and 16.1 have recently been obtained from a theoretical treatment of electrode processes at a smooth platinum electrode.[15]

*2. Isotopic equilibrium between the carboxyl hydrogen in acetic acid and hydrogen gas.* Isotopic equilibrium is readily established in the presence of a reduced platinum catalyst for the reactions:

$$HD + AcOH\ (1) \rightleftarrows H_2 + AcOD\ (1) \quad (1)$$

$$HT + AcOH\ (1) \rightleftarrows H_2 + AcOT\ (1) \quad (2)$$

At equilibrium, in reaction 1, the ratio of deuterium to protium in the carboxyl hydrogen is 3.83 times that in the hydrogen gas.[8] As was the case for the hydrogen-water equilibria described above, there is




I II

METHYL-3-$\alpha$ ACETOXY-$\Delta^{11}$ CHOLENATE

FIGURE 1

here a preferential distribution of the heavier isotope in the liquid phase. The preferential distribution factor in the case of reaction 2 is 6.85. These results aid in understanding gas-liquid isotopic equilibria prevailing when this system, including the catalyst, is used for hydrogenation purposes, in order to introduce deuterium or tritium into selected compounds. The reaction chosen for study was the hydrogenation of a steroid, methyl-3$\alpha$-acetoxy-$\Delta^{11}$, cholenate (I) to form the corresponding cholanate (II) (FIGURE 1). When the hydrogenating system contained only AcOD and deuterium gas, the product contained very close to 2.00 D atoms per molecule. Consequently it was assumed in the calculations that exactly two hydrogen atoms from the medium were incorporated during the hydrogenation. In this reaction, the acetic acid medium was present in large molar excess with respect to the hydrogen gas

Relative to the carboxyl hydrogen isotope content, the deuterium content of the atoms incorporated during hydrogenation is 3.3 times smaller while the tritium content is 5.4 times smaller.[8] The gas in these experiments was brought into isotopic thermodynamic equilibrium with the carboxyl hydrogen prior to the hydrogenation. The isotopic content of the carboxyl hydrogen dominates the result in these experiments, as shown by the observation that a factor of 6.0 is obtained when tank hydrogen gas is used at the start of the hydrogenation. It was concluded from these observations that isotopic exchange between dissolved hydrogen and carboxyl hydrogen or hydrogen ion is rapid relative to the reduction steps. The use of a different medium can yield different relative rates of exchange and reduction. For example, in the hydrogenation of maleic acid in aqueous solution using active platinum and palladium, it was found that the catalytic reduction is faster than the isotopic exchange between the hydrogen and water.[25]

That the heavier isotopes are preferentially incorporated into the steroid compound (FIGURE 1) by a factor of 1.10 and 1.26 for deuterium and tritium, respectively, *relative to the hydrogen gas composition*, is of interest, since the lighter isotopes are known to react more rapidly in many reactions that involve the rupture of a relatively strong valence bond. In a general treatment of the influence of the partition functions of the reacting molecules and the activated complex on relative isotopic rates, it was shown that the order of rate constants for reactions involving *free hydrogen atoms* as reactants would be $k_T > k_D > k_H$.[2] A similar conclusion had been reached after consideration of zero point energies for the initial and activated state.[23] These considerations would be applicable to the results of these experiments if it were assumed that the reacting hydrogen atoms are relatively loosely bound at the catalyst surface, so that isotopic zero point energy differences are small. The isotopic fractionation factor would also be affected by the isotopic exchange coefficient between hydrogen in the gas phase and the catalyst surface.

*3. Isotope selections effects.* The majority of isotope selections effects are consistent with the view that the rupture of a carbon-protium bond requires the smallest activation energy and consequently is accompanied by the fastest rate, if this step is rate-determining. Newton and Rollefson studied the relative rate of photochlorination of $CHCl_3$ and $CDCl_3$ in which the rate-determining step was considered to be $Cl + CHCl_3 \rightarrow CCl_3 + HCl$ and found that the reaction rate constant in the case of the $CHCl_3$ was approximately 4 times greater than for the $CDCl_3$ at room temperature.[22] The ratio $k_H/k_D$ for the oxidation of 2-propanol and 2-deuterio-2-propanol was in the range 5-8.[34] Benzaldehyde labeled with D in the aldehyde group was oxidized by potassium permanganate at a rate one-sixth as fast as ordinary benzaldehyde at pH 10.[35] In the oxidation of meth-

anol and ethanol by diazonium salts $k_H/k_T$ values of 5-7 were found.[21] The absence of isotopic fractionation in a reaction may, in some instances, be used in eliminating a particular step as rate-determining. The tritium content in the products of nitration of benzene, toluene, bromobenzene and naphthalene (each containing one labeled atomic position) could be explained only on the assumption that nitration at the C-T and C-H bonds takes place with the same speed.[20] It was therefore concluded that C-H bond breaking is not the rate-determining step in aromatic nitration. Identical rates of nitration for pentadeutero and ordinary nitrobenzene in concentrated sulfuric acid have recently been observed.[5] In agreement with the results above, it was concluded that breaking of the C-H bond did not influence the nitration step.

*II. Some* in Vitro *Reactions of Biochemical Interest*

The succinic acid obtained as a result of the hydrogenation of fumarate by deuterium gas in the presence of *Bacillus coli* had one fifth the theoretical content of heavy hydrogen based on the composition of the gas.[12] Several possible explanations were considered. In the presence of a succinic oxidase preparation, the rate of oxidation of the tetradeutero succinic acid was only 40 per cent of that for ordinary succinic acid while the rate of oxidation of the $\alpha, \alpha$ dideutero acid was about 70 per cent of that for the ordinary acid.[28] The activation energy was calculated from the slope of the log k *vs.* 1/T line. The difference in activation energy of 1450 ± 450 calories is of the order of magnitude of the zero point energy differences for C-H and C-D. These results are in accord with the hypothesis that the breaking of the C-H bond is an important rate-determining step in the overall process. Additional studies of this nature are needed to assist in a proper interpretation of the isotope-fractionation results. Some details of the mechanism of enzyme reactions may thereby be elucidated.

*III.* In Vivo *Studies Involving Hydrogen Isotope Fractionation.*

*1. Incorporation of hydrogen from the water medium in growing algae.* If an isotope effect did not exist, then deuterium and tritium in the aqueous medium would be incorporated at the same rate as ordinary hydrogen. As early as 1935, it had been observed that deuterium was incorporated into the nonexchangeable portion of growing algae to the extent of only 30 to 70 per cent of its concentration in the nutrient medium.[24] In these experiments the deuterium oxide concentrations ranged from 12 to 47 per cent. The uptake of tritium from an aqueous medium by the green alga, *Chlorella pyrenoidosa*, has recently been studied.[3] After a sufficient number of doublings the actual uptake of tritium is only about 50 per cent of that calculated for the case of no discrimination between the isotopes. The specific radioactivity of the medium was varied over a forty-fold range with no significant influence on this fractionation factor.

*2. Preferential incorporation of D relative to T into fatty acids (fats), cholesterol and glycogen following administration of water containing these isotopes.* The use of deuterium as a tracer in studying the transformation of precursor to product compounds has been a valuable tool in metabolic studies.[16, 26] When used properly as a label for the carbon atom to which it is attached, the interpretation of the results is usually straightforward. Where, however, the process studied involves the rupture of the carbon-hydrogen or deuterium bond in question, then there may occur a preferential utilization of one of the hydrogen isotopes. If the extent of the selection is relatively small, adequate qualitative and even semi-quantitative calculations can

still be made. If the extent of selection is large, than even the qualitative interpretation of mechanism may be faulty. Consequently, it is important to obtain some information concerning these selection factors. In the typical experiment in which a compound containing deuterium is administered to an animal, dilution with the same compound in normal isotopic abundance usually occurs. This makes it difficult to assay the extent of isotopic selection occurring between D and H. However, if precursor compounds containing both T and D are administered, a change in the ratio of T/D in the precursor to product will directly indicate the extent of selection for these isotopes. From these results, the extent of H:D and H:T fractionation may then be estimated.

Water containing both D and T was administered (in the drinking water) to rats. The isotopic content of the liver fatty acids was then measured and compared with the corresponding ratio in the water. (TABLE 1). The results in reference's 9 and 13 are in qualitative agreement and demonstrate a preferential utilization of the lighter isotope in the over-all conversion to this product. The results for total fat yield a smaller fractionation factor.[27] In similar experiments, the lighter isotope was preferentially utilized in the formation of liver glycogen by a factor of approximately ten per cent.[9] A factor of similar magnitude has recently been found for the liver total cholesterol after the administration of water labeled with deuterium and tritium.[10]

*3. Isotopic selection in the formation of the labile methyl group.* As a step in the direction of finding out whether tritium could be used as a satisfactory label for hydrogen, a study in the rat of methanols labeled with T, D and $C^{14}$ was undertaken.[30] It had already been found that, when methanols containing $C^{14}H_3OH$ and $CD_3OH$ was administered, approximately one-fourth to one-third of the deuterium appeared in the methyl group of choline relative to the $C^{14}$ content of this group.[3] The utilization of methanol for methyl synthesis was therefore interpreted as occurring through an oxidation and subsequent reduction to the labile methyl group. This was in contrast with the results that had been obtained following the administration of methionine labeled with $C^{14}$ and D in the methyl group.[17] In the latter experiment, the deuterium to $C^{14}$ ratio in the choline methyl group was the same as in the methyl group of the administered methionine.

The isotopic analyses of the labile methyl groups of choline and creatine biosynthesized in the rats following administration of methanol containing $C^{14}H_3OH$, $CD_3OH$ and $CH_2TOH$ are listed in TABLE 2.[30] The results on the comparison of the ratio of deuterium to $C^{14}$ in the choline and the administered methanol (column 3) are in fairly good agreement with those obtained

TABLE 1

FACTOR FOR PREFERENTIAL INCORPORATION OF DEUTERIUM RELATIVE TO TRITIUM INTO FATTY ACIDS FOLLOWING ADMINISTRATION OF WATER CONTAINING D AND T IN THE RAT.

| Duration of experiment (days) | Atom % excess deuterium in water | Organ | Factor | Reference |
|---|---|---|---|---|
| >14 | 2.5 | Mammary gland | 1.2 | Glascock and Dunscombe[13] |
| 1-4 | <2 | Liver | 1.17-1.19 | Eidinoff *et al.*[9] |
| 10 | 5 | (Bound fat) | 1.06 | Thompson and Ballou[27] |

TABLE 2

COMPARISON OF ISOTOPIC CONTENT OF LABILE METHYL GROUPS WITH ADMINISTERED METHANOL

| Rat | Compound | $F_D$* | $F_T$* | $F_T/F_D$ |
|---|---|---|---|---|
| J | TMCP† | 0.22 | 0.75 | 3.4 |
| | MCP‡ | .36 | 1.05 | 2.9 |
| F | TMCP | .22 | 0.69 | 3.1 |
| | MCP | .28 | 0.86 | 3.1 |

* $F_D$ or $F_T$ are the fractions obtained by dividing the deuterium to $C^{14}$ ratio or the tritium to $C^{14}$ ratio, respectively, in the methyl group of the isolated compounds by the D:$C^{14}$ or T:$C^{14}$ in the methyl group of the administered methanol.

† TMCP = trimethylamine chloroplatinate.

‡ MCP = methylamine chloroplatinate.

in the earlier experiments with methanol labeled with $C^{14}$ and deuterium.[31] In these earlier experiments, the ratio of deuterium to $C^{14}$ in the choline was 25 to 31 per cent of the ratio in the methyl group of the administered methanol. However, the ratio of tritium to $C^{14}$ in the methyl group of the choline was 69 to 75 per cent of that in the methyl group of the administered methanol (column 4). The $F_T/F_D$ values in the last column are a direct measure of the preferential retention of the carbon-tritium bond in the *in vivo* transformation of the methyl group in methanol to the labile methyl group. If the isotopes of hydrogen were completely indistinguishable, then the results in the last column would be unity. These results demonstrate directly a selection between deuterium and tritium in the over-all biological transformation studied, and furnish indirect evidence that selection effects of similar magnitude are taking place between protium and deuterium.

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## THE THEORY OF ISOTOPIC FRACTIONATION IN KINETIC AND EQUILIBRIUM SYSTEMS (ABSTRACT)*

*By* JACOB BIGELEISEN†

The fractionation of isotopes by exchange reactions in equilibrium systems is a pure quantum effect. This type of fractionation arises from the fact that isotopic exchange reactions are accompanied by a small heat of reaction, which results from differences in zero point energies. In some cases exact calculations can be made from spectroscopic data, yielding results in complete agreement with experiment.

Since the equilibrium constant for an isotopic exchange reaction depends on the structures of the molecules in the exchange reaction, such studies can be used to evaluate structural parameters. Examples are given concerning structural data of $NH_3$ and $H_3O^+$ obtained from deuterium exchange equilibria.

Isotopes react at different rates because they affect two of the factors which control the reaction rate. One is the activation energy and arises from the zero point energy effect. This is a quantum phenomenon. The second, a classical effect, is the effect of isotopic substitution on the frequency of dissociation of activated complexes. The frequency effect accounts for the fact that the isotopic fractionation factors in kinetic systems are generally larger than those in equilibrium systems.

* This paper, the second of two papers in a symposium on Fractionation Effects Involving the Isotopes of Hydrogen, was presented at a meeting of the Section on November 10, 1953.

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## *SECTION OF PSYCHOLOGY*

# SOME OBSERVATIONS ON PSYCHOLOGY IN JAPAN*

*By* C. H. GRAHAM†

This paper presents a survey of Japanese psychology, first from an historical point of view, and second, from the point of view of analyzing some problems that confront its modern representatives. The occasion for organizing the paper arose in the summer of 1952 when Professor Sato, of Kyoto University, and the writer were members of the Seminar in Experimental Psychology, one of the Kyoto Seminars in American Studies, under the auspices of Kyoto University, Doshisha University, the University of Illinois, and the Rockefeller Foundation.

The first part of the paper deals with the historical background of psychology in Japan. The historical sequence is broken up into three parts: from the beginnings of psychology (about 1890) until 1926; from 1926 through World War II; and from the end of World War II to the present.

### *History of Psychology in Japan*

*Psychology until 1926.* Western psychology was introduced into Japan about 1890 against an ancient background of Indian and Buddhist philosophy.

Yujiro Motora (1858-1912) and Matataro Matsumoto (1865-1943) were the principal founders of Japanese psychology. Motora was the first professor of psychology in Tokyo University. He had a philosophic mind and tried to establish a system of psychology. Matsumoto was primarily an experimentalist. He survived Motora for thirty-six years, and most of the older leaders of Japanese psychology are his students.

The first twenty-five years of the twentieth century constituted an orientation period in the methods of psychological research. The work was carried out primarily in the psychological laboratories of Tokyo and Kyoto Universities.

With the advent of better economic conditions after World War I the number of educational institutions increased greatly. Departments of psychology were established in 12 of the leading universities and, in addition, many psychologists obtained positions in the Kotogakko (essentially junior colleges).

It is not surprising that the increase in Japanese educational facilities was accompanied by a quickened interest in educational psychology, an interest that was becoming strongly established by World War I. In general, however, it may be said that areas of psychology other than educational and experimental received little attention at this time.

*From 1926 through World War II.* The beginning of the Showa Era (1926) marked the development of some important trends in Japanese psychology. At that time the influence of Gestalt psychology was beginning to be felt; the Japanese Psychological Association was established, as was the new series of the *Japanese Journal of Psychology*, and the number of graduates in psychology was showing a remarkable increase due to the expanded system of higher education that occurred after World War I.

Onoshima and Sakuma, who studied in Berlin University, were the chief expositors of Gestalt psychology. It can safely be said that Gestalt theory has colored the thought of most Japanese psychologists from about 1926 to World War II (and even now exerts a powerful influence).

*Experimental psychology.* In the years between 1926 and 1939, the study of perception progressed at a quickened pace, usually in a line dictated by Gestalt psychology. In particular, a number of investi-

*This paper was presented at a meeting of the Section on November 16, 1953.

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gations were carried out on the visual constancies, depth perception and movement that are, unfortunately, not so well known in western countries as they should be. Little work was done on other sensory systems.

The field of human learning was not so intensively investigated from 1926 to 1939 as was the area of perception. Studies that were made were done within the general framework of Gestalt psychology.

Little or no work was done in the field of animal learning before the end of World War II. Hayashi, a student of Pavlov, pioneered in the study of conditioning but, except for him, little was done until after the war.

*Personality.* In the decade of the 30's, German characterology was introduced into Japan and some experiments were performed within the framework of this viewpoint. These experiments constituted the first systematic work in the field of personality. For example, Uchida devised a continuous addition test, essentially a modification of Krapelin's work. During this period, investigators under the guidance of Chiba, studied personality differences among Manchurian races and, in another direction, Masaki and Yoga studied personality from the viewpoint of educational psychology.

*Educational and clinical psychology.* Educational psychology showed considerable progress between 1926 and World War II. Studies of development, learning, personality, work and fatigue increased greatly in number and had direct influences on education despite the prevalence of a strong German influence, generally unsympathetic to developmental studies.

In the 1930's psychologists, child psychologists, and educationists formed groups to study clinical and abnormal child psychology. Their studies constituted the beginnings of clinical psychology in Japan. A few clinics were established in prisons, courts, and reformatories under the Japanese Judicial Department and in guidance centers set up by the Welfare Department. The work of the early clinicians overlapped the work of the educational psychologists, but a difference did exist in one respect: The former extended their methods to meet the requirements of evaluating abnormal children and criminals. In a few cases the clinicians adopted psychoanalytic methods.

*Industrial psychology.* Industrial psychology, always at a disadvantage in Japan because of ever-present overpopulation and its consequence, cheap labor, did not progress so rapidly during the 1930's as some other areas of psychology. It has been stated that, during World War I, attention was paid to industrial psychology, and some systematic studies of the labor problems were made subsequent to 1926 by members of the Kurashiki Institute of the Psychology of Labor, founded in 1921 by Ohara. By the middle of the 1930's the Ministry of Welfare and the Government Railways employed a number of psychologists for vocational guidance and the promotion of efficiency of labor.

*Social psychology.* Wundt's folk psychology and McDougall's social psychology were studied at an early date by Japanese psychologists, but research in the social area did not develop for a long time. The measurement of attitude was first undertaken by Koga about 1934 and problems of race differences and group psychology interested psychologists from about 1931 to World War II.

*The general trend: 1926 through World War II.* Some general trends are evident from 1926 through World War II. During this period, Japanese psychologists were extremely self-conscious and self-critical. Matsumoto, President of the Japanese Psychological Association in 1933, discussed what he considered to be the three weak points of Japanese psychology: (1) Lack of

communication among psychologists. (2) Excessive use of group methods involving questionnaires. (3) Excessive speed in adopting new, and probably, transient western psychological concepts.

The years near 1935 were the high watermark of Japanese psychology before World War II. The number of psychological journals (about eight) was at a peak and psychologists were very active. Then came the precipitous decline. Psychological research in Japan was severely reduced at the time of the outbreak of the China-Japan Conflict in 1937 and its later development into the Pacific War in 1941. Several journals could not be continued, and at the end of the war even the *Japanese Journal of Psychology* could not be published regularly. Psychological research was almost completely interrupted, and the number of graduate students dwindled to essentially zero.

*From the End of World War II to the Present.* After World War II a great number of changes took place in Japanese life, many of them attributable to the Occupation. Among other things, the changes included a new Constitution and a proposed renovation of the Japanese educational system. At this time, the number of graduate students in psychology increased considerably above the prewar level, the Japanese Psychological Association increased its membership greatly, and its meetings came to be held annually.

*Experimental psychology.* Inferest has increased since the war in psychophysiological problems. Experiments on movement are being carried on, and they, with current studies of size and shape discrimination, maintain ties with the earlier Gestalt influence, now somewhat mixed with behavioristic tendencies. A latent behavioristic influence has now become fairly well established, manifested particularly by such men as Imada and Kotake at Kwansei Gukuin University. The topic of figural after-effects is being studied with considerable enthusiam and the Japanese researches are being done effectively. Other areas of visual perception are also being examined.

The greatly increased interest in animal experimentation is of major postwar development. It is worth noting that a Society for the Study of Behavior Theory has been established by psychologists of the Universities of Kyoto and Osaka. Considerable interest also exists in problems of human learning, but little work is being performed on motor skills.

*Personality.* Research on personality *per se* has not expanded after the war to the same degree as some other areas of interest. However, it is important to observe that many investigators are at work on experiments that are closely related to this area, for example, those that deal with the influence of motives on discrimination.

*Developmental psychology and measurement.* The reform of the Japanese educational system, instituted by the Occupation, involves an important program of teacher education and re-education. The re-education program has placed great stress on the data of educational psychology and, under the new system, many school teachers have been instructed in this area.

Recently several books on the developmental aspects of educational psychology have been translated into Japanese and several books have been written on child psychology with an essentially educational emphasis, one for example, by Kubo, who owes much to Charlotte Buhler, and another by Tatano, who is in the tradition of Piaget.

*Clinical psychology.* The status of clinical psychology in Japan has changed greatly in the postwar period. The name "clinical psychology" is now in common use and the Society of Clinical Psychology has been formed. A journal of clinical psychology has recently been established and now, for the first time, psychoanalytic theories are becoming common subjects of

discussion. In addition, some truly Japanese methods, such as Worita's, which are based upon a synthesis of Zen doctrines and western studies, are regarded favorably.

*Industrial psychology.* Although industrial psychology in Japan has not developed to the same degree as in the west, it has nevertheless been shown that a basic core of subject matter, technique, and practice did exist prior to World War II. Since that war, labor problems have received more attention than previously. Several prefectures have established research institutions for work in this area and a few universities have set up chairs of industrial psychology and vocational guidance.

*Social psychology.* After the war many social problems forced themselves upon the attention of psychologists. At the same time, the theories and experiments of social psychologists in America flowed into Japan and stimulated the Japanese workers. In consequence, work in social psychology is now being carried on at an accelerated pace.

At first, after the war, considerable effort was spent on the analysis of social phenomena that occurred during and after hostilities. Later, attention was paid to problems of group dynamics. More recently, problems of social perception and the measurement of attitude became a central problem not only for social psychologists but for many others as well. Most recently, psychologists have become interested in systematic problems. Several research groups have been formed, for example, the Society for Research in Behavior Mechanisms (at Tokyo), the Association for Group Dynamics (at Kyushu), and the Society for Youth Work (at Tokyo).

### *An Overview*

Our survey points to the fact that psychology got off to a slow start in Japan. During the first twenty-five years of the twentieth century psychology did not develop surely and rapidly. By 1935 it gave promise, not only of becoming stronger but also of becoming more diversified. Unfortunately, this favorable pattern of growth was utterly destroyed by the events that led to the Chinese Conflict and World War II. Following the war, Japanese psychology underwent the readjustment common to all areas of Japanese life. At the present time, it shows strong signs of revival at a new and stronger level of adaptation. The future success of Japanese psychology may, to a large extent, be determined by how well it defeats or adjusts to unfavorable problems, problems that arise from the conflict between new tendencies and resistant realities.

What, we may ask, are some of these problems and in what areas do they appear?

*Current problems of Japanese psychology training.* Under the Occupation (1945-1952) a strong effort was made to remodel the education system of Japan.

So far as concerns graduate work in psychology, it is sufficient to say that the "new" system proposed by Occupation authorities specified that 30 credits of course work be required in the first year of graduate study. These credits were to be accumulated in the areas of experimental, clinical, social, developmental, educational, and certain optional areas. (As of the summer of 1952, the "new" program did not seem to be receiving enthusiastic support. The main argument against it is that it is expensive both for the university and for the student.)

Under the "old" prewar system, which is still strongly supported, few examinations were given during a man's graduate work and the requirements for reports on research varied from university to university. The major professor decided when a man should finally stop his training. A doctorate degree was granted, if it were granted at all, usually after many years of professional work. It was based upon a defense of the thesis.

What opinions do Japanese psychologists have about ways of improving graduate training? The following proposals were

frequently encountered and seem to represent the prevailing views of the Seminar participants:

(1) Establish a program leading to a terminal doctor's degree that would be obtainable after a reasonable period of graduate study.

(2) Specify more appropriate formal prerequisites for graduate training in each area of psychology.

(3) Provide more adequately supervised work in the laboratory and in the clinic.

(4) Improve facilities for research and graduate training.

(5) Place psychology in some other faculty than the Faculty of Literature.

Some of the proposals may soon be carried out under any circumstances. For one thing, psychology may vacate its place in the Faculty of Literature. Certain government officials seem to favor the reclassification of psychology, but others do not, probably on the grounds that, as a recognized laboratory and clinical discipline, it will become more expensive. In any case, psychologists themselves, by establishing the objective of producing students thoroughly but broadly trained in subject matter and skillful in the use of research techniques, will go a long way to improve the status of graduate training.

*Professional opportunities.* The lot of a psychologist in Japan has many desirable features but, financially, it is no more attractive than that of many other academic people. For example, a full professor may earn about $100 per month, a salary that is quite in line with prevailing salaries. Nevertheless, academic people are certainly not rich!

Another main flaw in the professional picture lies in the fact that psychology does not have a high status in Japan. It has not been exploited by government, education, and industry, as has been the case in the west, and few positions exist for psychologists outside of universities.

*Problems of communication.* Psychologists in Japan feel isolated and, in fact, they have been isolated, not only from western psychologists but, to a greater degree than is desirable, from each other.

The isolation from western psychologists is attributable to a great many factors, among which geographical position plays a minor role. The lack of contact with the western world that existed from 1937 to 1945 had its undesirable effects, and the Occupation years (1945-52) that followed constituted a period of readjustment that did not provide the best circumstances for scientific give and take.

More than any other factor, however, the barrier of language has served to confirm the isolation of the Japanese. Despite the fact that Japanese training in foreign languages is good, the training of western workers in Japanese is bad. It has been the lot of Japanese psychologists since their beginnings to do research which remains little known to the outside world. In consequence, Japanese psychologists have had to depend solely on the approval of their fellow countrymen.

Certain devices for increasing communication might be tentatively recommended without committment as to their ultimate value. For example, Japanese psychologists could be encouraged and helped to publish in English, as many do now in the sense that they provide long English summaries of their articles. Other devices such as intercultural seminars, exchange scholarships, exchange professorships, intercultural institutions, *etc.*, might produce more intimate scientific relations. Certainly programs in these directions should be encouraged.

### *Concluding Remarks*

Psychology in Japan has had to cope with a number of unique problems. Some of the problems remain and new ones may arise. Nevertheless, morale among Japanese psychologists is high; they are ready to help themselves. The outlook for psychology in this easternmost country is probably as good or better than it ever was.

## *SECTION OF MATHEMATICS AND ENGINEERING*

# SOME SELECTED QUICK AND EASY METHODS OF STATISTICAL ANALYSIS*

*By* JOHN W. TUKEY†

### *Introduction*

The methods to be discussed are quick and easy but do not, and need not, claim to be definitive. For every set of experimental or observational data which requires or deserves definitive analysis, there are many which deserve some reasonable degree of analysis. All too often they do not get any analysis at all!

By analysis, what do we mean? In the typical experiment, (and we shall always include observational programs as experiments), some causes of fluctuation are *visible*—are allowed to show themselves—usually by contributing to differences among the experimental results. Other causes of fluctuation are concealed from the experimenter or his critic alike, usually because a choice or an operation is performed only once (this is now frequently referred to as duplicity, in analogy to the classically conducted duplicate analysis of the chemist). Other causes of error cannot show in the experiment because they are constant or because they vary slowly; these are *systematic* errors. Systematic errors are held in check or reduced by the application of subject matter knowledge, whether it be science, engineering, market research, or accounting. Fluctuations are made visible (removed from concealment, with a consequent reduction in duplicity) or made harmless by the combination of subject matter knowledge and the techniques of statistical design of experiment. It remains for statistical analysis to assess the likely influence of the *visible* fluctuations on the apparent results of the experiment, (to assert that the results indicated so-and-so within such-and-such allowances, *except* for the effects of concealed fluctuations or systematic errors) and to indicate that the results might well be this accurate and are surely no more accurate.

### *A Single Group of Measurements*

Surely the simplest experiment is one in which a single quantity is measured a number of times. Thus Davies[1] has reported the yields of naphthalene black 12B, expressed in grams of standard color, for 5 measurements on a certain sample of H-acid (a well-known dye intermediate). The observations and their total and range were

| | |
|---|---|
| | 1510 |
| | 1550 |
| | 1560 |
| | 1595 |
| | 1605 |
| Total | 7820 |
| Range | 95 = 1605 − 1510 |

What limits on the total are indicated by the overt fluctuations in the results? Reference to TABLE 1 shows that for a 5 per cent error rate we should multiply the range of 5 observations by 2.54, obtaining 242 in this example. The result is

$$7820 \pm 242 = \text{between } 7578 \text{ and } 8062$$

for the total. The mean of the 5 determinations is 1/5 of this, and is thus between 1516 and 1624 with a 5 per cent error rate.

What is this 5 per cent error rate? Its meaning is the following: Hold the systematic errors and the errors of duplicity constant. Repeat the measurement very many times. Call the average result the *determinand*. (This is what we were actually determining, the quantity to which our 5 measurements are relevant! If systematic errors and errors of duplicity are small, it

* This paper was presented at a meeting of the Section on November 20, 1953. It was prepared in connection with research sponsored by the Office of Naval Research.

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TABLE 1

VALUES OF [ALLOWANCE FOR TOTALS/RANGE] FOR A SINGLE GROUP OF MEASUREMENTS.

| *Number of measurements* | *Ratio of allowance to range* |
|---|---|
| 2 | 12.71 |
| 3 | 3.91 |
| 4 | 2.87 |
| 5 | 2.54 |
| 6 | 2.39 |
| 7 | 2.33 |
| 8 | 2.30 |
| 9 | 2.30 |
| 10 | 2.30 |
| 11 | 2.32 |
| 12 | 2.33 |
| 13 | 2.35 |
| 14 | 2.38 |
| 15 | 2.40 |
| 16 | 2.42 |
| 17 | 2.45 |
| 18 | 2.47 |
| 19 | 2.49 |
| 20 | 2.50 |

is nearly what we wanted to measure!) Call the concluding statement of our analysis, in the example "between 1516 and 1624," correct if the interval includes the determinand; an error if it fails to include the determinand. The 5 per cent error rate means that the analysis of 5 per cent of all experiments will be errors; *i.e.*, 5 per cent of such statements will fail to make adequate allowance for the effects of visible fluctuations.

There is nothing sacred about 5 per cent; it is merely a convenient standard. We can operate similar procedures at any error rate we choose. However, the 5 per cent interval will often serve as a basis for rough but adequate estimates of the intervals for other error rates. For simplicity, we shall stick to 5 per cent in this account.

Just as we would all criticize the experimenter with 5 measurements who did not at least total the 5 measurements and divide by 5 to find their mean (or at least summarize the 5 values by some other typical value), so we should, in the writer's opinion, also criticize the experimenter who does not at least find the range of his observations and, by the simple calculation just exhibited (or by some other procedure), set some limits on his mean (or other typical value). If the mean is worth calculating at all, then it must be worth while setting some rough limits on the determinand to which it refers. We know that visible fluctuations are always present. How could we avoid indicating the apparent size of their effect in our summary of the measurements? Clearly "between 1516 and 1624" is a more useful summary than "apparently 1564"!

I hope that these remarks are regarded by many as obvious and unnecessary as applied to this example, for then how can they fail to agree that we should behave similarly in more complex situations! (And in these they may well have to break away from old habits.)

### *Two Groups of Measurements*

Actually the sample of H-acid just discussed was not alone. We may take from Davies[1] the following:

| | *Sample A* | *Sample B* |
|---|---|---|
| | 1510 | 1445 |
| | 1550 | 1450 |
| | 1560 | 1455 |
| | 1595 | 1480 |
| | 1605 | 1520 |
| Total | 7820 | 7350 |
| Range | 95 | 75 |

Now the total range is 95 + 75 = 170, and the factor for comparison of the two totals can be taken from TABLE 2. It is 1.53, so that the allowance is ±260, and the difference in totals is

$$7820 - 7350 \pm 260 = 470 \pm 260 =$$
between 210 and 730

Dividing by 5, we find a difference in means of between 42 and 146, again with a 5 per cent error rate. This means that the average of many determinations under these particular conditions on sample A should be 42 gms of color to 146 gms of color higher than the corresponding average for sample B.

Since these two samples of H-acid probably came from two batches produced under similar conditions, it is likely that concealed and systematic errors were quite similar for the two samples. The allowance for the effects of visible fluctuations *may* do quite well in allowing for visible and concealed fluctuations and systematic errors. (Of course, it may not. One sample may contain a peculiar contaminant which causes unusual systematic errors.) In general, however, comparisons are considerably less affected by systematic errors than are absolute measurements, and the results of analysis are correspondingly more useful.

Techniques for putting limits on a comparison of this sort have been available for many years, yet too many users of my acquaintance have learned only to test whether the difference might be zero. Blame for this state of affairs must rest on the statisticians.

### *Several Groups of Measurements*

*Example 1. The balanced single classification.* Let us return to Davies and the H-acids—let us return to face the fact that there were six samples, not just two! Until recently, the only techniques of analysis available were ones to test whether all samples might be alike—we asked only whether we must confess that there must be some differences! (If you agreed to the necessity of limits with one sample or two samples, how can you fail to ask for some sort of limits for six samples?) Recently, methods have been made available for the setting of limits in such situations. There are both definitive methods for occasional use, and quick and easy methods for routine use. These methods differ among themselves as to their detailed aims and statement of error rates—we shall present only one brand, trying to specify its properties clearly.

We have a *family* of six samples of H-acid to be compared, as a minimum we wish to compare each sample with every other sample, a total of ½ (5)6 = 15 comparisons. We wish to make statements about all 15 comparisons, and to do this in such a way that the error rate is known. Like other rates, this error rate has both a numerator and denominator. We must concern ourselves with both. The brand of technique presented here is deliberately conservative, and chooses for the denominator the number of families involved and for the numerator the number of families in which *one or more* statements about the differences between determinands is in error. In other words, we wish to set limits on all differences between determinands so that *all* these limits will be correct in 95 per cent of all families. (This is a conservative standard, to be sure, but no more conservative than that adopted in F-testing simple analyses of variance.)

Having decided what to do, and calculated the necessary tables, we have only to proceed in an entirely similar way. Davies provides the measurements and we supply totals and ranges with the following results:

| Sample: | *1* | *2* | *3* | *4* | *5* | *6* |
|---|---|---|---|---|---|---|
| Color (gms): | 1440 | 1490 | 1510 | 1440 | 1515 | 1445 |
| | 1440 | 1495 | 1550 | 1445 | 1595 | 1450 |
| | 1520 | 1540 | 1560 | 1465 | 1625 | 1455 |
| | 1545 | 1555 | 1595 | 1545 | 1630 | 1480 |
| | 1580 | 1560 | 1605 | 1595 | 1635 | 1520 |
| Total: | 7525 | 7640 | 7820 | 7490 | 8000 | 7350 |
| Range: | 140 | 70 | 95 | 155 | 120 | 75 |

The sum of the ranges is

$$140 + 70 + 95 + 155 + 120 + 75 = 655$$

and from TABLE 2 we find a factor of .70, so that the allowance is ±(.70)(655) = ±457. Dividing totals and allowance by 5, we obtain the summary:

| *Sample* | *Mean color (gms)* |
|---|---|
| 1 | 1550 |
| 2 | 1528 |
| 3 | 1564 |
| 4 | 1498 |
| 5 | 1600 |
| 6 | 1470 |
| WSD | ±91 |

TABLE 2

VALUES OF [(WSD FOR TOTALS) / (TOTAL RANGE)] FOR A BALANCED SINGLE CLASSIFICATION AND AN ERROR RATE OF 5 PER CENT FAMILYWISE

number of groups = number of ranges = number of determinations

number of observations per group, range or determination

| | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 2 | 3.43 | 2.37 | 1.78 | 1.40 | 1.16 | 1.00 | .87 | .78 | .70 | .66 | .63 | .58 | .50 | .47 | .44 | .42 | .40 | .38 | .36 |
| 3 | 1.91 | 1.44 | 1.13 | .94 | .80 | .70 | .62 | .56 | .51 | .47 | .43 | .40 | .38 | .36 | .33 | .32 | .30 | .29 | .27 |
| 4 | 1.63 | 1.25 | 1.01 | .84 | .72 | .63 | .57 | .51 | .47 | .43 | .40 | .37 | .35 | .33 | .31 | .29 | .28 | .27 | .25 |
| 5 | 1.53 | 1.19 | .96 | .81 | .70 | .61 | .55 | .50 | .45 | .42 | .39 | .36 | .34 | .32 | .30 | .29 | .27 | .26 | .25 |
| 6 | 1.50 | 1.18 | .95 | .80 | .69 | .61 | .55 | .49 | .45 | .42 | .39 | .36 | .34 | .32 | .30 | .29 | .27 | .26 | .25 |
| 7 | 1.49 | 1.17 | .95 | .80 | .69 | .61 | .55 | .50 | .45 | .42 | .39 | .36 | .34 | .32 | .30 | .29 | .28 | .26 | .25 |
| 8 | 1.49 | 1.17 | .96 | .81 | .70 | .62 | .55 | .50 | .46 | .42 | .39 | .37 | .35 | .33 | .31 | .29 | .26 | .27 | .25 |
| 9 | 1.50 | 1.18 | .97 | .82 | .71 | .62 | .56 | .51 | .47 | .43 | .40 | .37 | .35 | .33 | .31 | .30 | .28 | .27 | .26 |
| 10 | 1.52 | 1.20 | .98 | .83 | .72 | .63 | .57 | .52 | .47 | .44 | .41 | .38 | .35 | .34 | .32 | .30 | .29 | .27 | .26 |
| 11 | 1.54 | 1.21 | .99 | .84 | .73 | .64 | .58 | .52 | .48 | .44 | .41 | .38 | .36 | .34 | .32 | .31 | .29 | .28 | .27 |
| 12 | 1.56 | 1.23 | 1.00 | .85 | .74 | .65 | .59 | .53 | .49 | .45 | .42 | .39 | .37 | .35 | .33 | .31 | .30 | .28 | .27 |
| 13 | 1.58 | 1.25 | 1.02 | .86 | .75 | .66 | .59 | .54 | .49 | .46 | .42 | .40 | .37 | .35 | .33 | .32 | .30 | .29 | .27 |
| 14 | 1.60 | 1.26 | 1.03 | .87 | .76 | .67 | .60 | .55 | .50 | .46 | .43 | .40 | .38 | .36 | .34 | .32 | .31 | .29 | .28 |
| 15 | 1.62 | 1.28 | 1.05 | .89 | .77 | .68 | .61 | .56 | .51 | .47 | .44 | .41 | .38 | .36 | .34 | .33 | .31 | .30 | .28 |
| 16 | 1.64 | 1.30 | 1.06 | .90 | .78 | .69 | .62 | .56 | .52 | .48 | .44 | .41 | .39 | .37 | .35 | .33 | .31 | .30 | .29 |
| 17 | 1.66 | 1.31 | 1.08 | .91 | .79 | .70 | .63 | .57 | .52 | .48 | .45 | .42 | .39 | .37 | .35 | .33 | .32 | .30 | .29 |
| 18 | 1.68 | 1.33 | 1.09 | .92 | .80 | .71 | .64 | .58 | .53 | .49 | .46 | .43 | .40 | .38 | .36 | .34 | .32 | .31 | .30 |
| 19 | 1.70 | 1.34 | 1.10 | .93 | .81 | .72 | .65 | .59 | .54 | .50 | .46 | .43 | .40 | .38 | .36 | .34 | .33 | .31 | .30 |
| 20 | 1.72 | 1.36 | 1.11 | .95 | .82 | .73 | .65 | .59 | .54 | .50 | .47 | .44 | .41 | .39 | .37 | .35 | .33 | .32 | .30 |

(WSD stands for the type of allowance here calculated, and may be remembered as the "wholly significant difference.") The difference between sample 5 and sample 4 is thus stated to be

$$1600 - 1498 \pm 91 = 102 \pm 91 = 11 \text{ to } 193 \text{ gms},$$

while that between sample 5 and sample 6 is stated to be

$$1600 - 1470 \pm 91 = 130 \pm 91 = 39 \text{ to } 231 \text{ gms},$$

that between sample 1 and sample 2 to be

$$1550 - 1528 \pm 91 = -69 \text{ to } 113 \text{ gms},$$

and so on for the other 12 comparisons.

Since our standard of success is that all statements must be correct, we can combine these statements freely without increasing our error rate. Thus, for example, the difference between sample 5 and the *higher* of samples 4 and 6 is also

11 to 193 gms of color,

while the difference between sample 5 and *either* of samples 4 and 6 is

11 to 231 gms of color.

Moreover, sample 5 may be as much as

$$36 + 91 = 127 \text{ gms of color}$$

better than all the others, or as much as

$$-(36 - 91) = 55 \text{ gms of color}$$

worse than the best of the others.

These combinations need not be in terms of the "the higher of," "the lower of," "either of," "the highest of," "the lowest of," or "any of," they may also be in terms of such linear combinations as

$$x_1 - \frac{x_2 + x_3}{2} = 1550 - \frac{1528 + 1564}{2} \pm 91 =$$

$$4 \pm 91 = -87 \text{ to } +95,$$

or

$$0.3x_1 + 0.4x_3 + 0.3x_5 - 0.8x_2 - 0.2x_4 =$$

$$1570.6 - 1520.8 \pm 91 = 50 \pm 91 = -41 \text{ to } +141,$$

So long as the sum of the positive coefficients equals unity, and the sum of the negative coefficients also equals unity, the same conclusions are made available by specifying the 6 means and the WSD.

### *Sources*

The methods for one and two groups are arithmetically streamlined versions of those developed by E. Lord[4] in 1947.

TABLE 3

VALUES OF [(WSD FOR TOTALS) / (TOTAL RANGE OF DIFFERENCES)] FOR A BALANCED DOUBLE CLASSIFICATION AND AN ERROR RATE OF 5 PER CENT FAMILYWISE. (UPPER ENTRIES FOR TOTALS A, LOWER ENTRIES FOR TOTALS B)

Number of ranges = Number of totals A

| Number of differences per range = Number of totals B | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 2 | 6.36 | 2.17 | 1.53 | 1.17 | .96 | .80 | .68 | .62 | .54 | .49 | .45 | .41 | .38 | .36 | .34 | .31 | .30 | .28 | .27 |
| | 6.36 | 1.95 | 1.40 | 1.12 | .96 | .84 | .76 | .70 | .65 | .61 | .58 | .55 | .52 | .50 | .48 | .46 | .45 | .44 | .42 |
| 3 | 1.95 | 1.17 | .90 | .73 | .62 | .54 | .48 | .42 | .38 | .35 | .32 | .30 | .28 | .26 | .25 | .23 | .22 | .21 | .20 |
| | 2.17 | 1.17 | .91 | .77 | .68 | .61 | .56 | .52 | .49 | .46 | .44 | .42 | .40 | .38 | .37 | .36 | .35 | .34 | .33 |
| 4 | 1.45 | .96 | .76 | .63 | .54 | .47 | .42 | .38 | .34 | .32 | .29 | .27 | .25 | .24 | .23 | .21 | .20 | .19 | .18 |
| | 1.56 | .94 | .76 | .66 | .58 | .53 | .49 | .45 | .43 | .40 | .39 | .37 | .35 | .34 | .33 | .32 | .31 | .30 | .29 |
| 5 | 1.27 | .90 | .72 | .60 | .51 | .45 | .40 | .36 | .33 | .30 | .28 | .26 | .25 | .23 | .22 | .21 | .20 | .19 | .18 |
| | 1.29 | .84 | .69 | .60 | .53 | .49 | .45 | .42 | .40 | .38 | .36 | .34 | .33 | .32 | .31 | .30 | .29 | .28 | .27 |
| 6 | 1.18 | .87 | .70 | .58 | .50 | .44 | .39 | .36 | .33 | .30 | .28 | .26 | .24 | .23 | .22 | .21 | .20 | .19 | .18 |
| | 1.14 | .78 | .64 | .56 | .50 | .46 | .43 | .40 | .38 | .36 | .34 | .33 | .31 | .30 | .29 | .28 | .27 | .27 | .26 |
| 7 | 1.16 | .85 | .69 | .58 | .50 | .44 | .39 | .36 | .33 | .30 | .28 | .26 | .24 | .23 | .22 | .21 | .20 | .19 | .18 |
| | 1.07 | .74 | .61 | .54 | .48 | .44 | .41 | .38 | .36 | .34 | .33 | .31 | .30 | .29 | .28 | .27 | .26 | .26 | .25 |
| 8 | 1.15 | .85 | .69 | .58 | .50 | .44 | .40 | .36 | .33 | .30 | .28 | .26 | .25 | .23 | .22 | .21 | .20 | .19 | .18 |
| | 1.00 | .71 | .59 | .52 | .47 | .43 | .40 | .37 | .35 | .33 | .32 | .30 | .29 | .28 | .27 | .26 | .26 | .25 | .24 |
| 9 | 1.14 | .85 | .69 | .58 | .50 | .45 | .40 | .36 | .33 | .31 | .28 | .27 | .25 | .24 | .22 | .21 | .20 | .19 | .18 |
| | .96 | .68 | .57 | .50 | .45 | .42 | .39 | .36 | .34 | .33 | .31 | .30 | .29 | .28 | .27 | .26 | .25 | .24 | .24 |
| 10 | 1.15 | .86 | .70 | .59 | .51 | .45 | .40 | .37 | .34 | .31 | .29 | .27 | .25 | .24 | .23 | .21 | .20 | .19 | .19 |
| | .93 | .66 | .56 | .49 | .44 | .41 | .38 | .36 | .34 | .32 | .31 | .29 | .28 | .27 | .26 | .25 | .25 | .24 | .23 |
| 11 | 1.15 | .87 | .71 | .60 | .52 | .46 | .41 | .37 | .34 | .31 | .29 | .27 | .26 | .24 | .23 | .22 | .21 | .20 | .19 |
| | .91 | .65 | .55 | .48 | .43 | .40 | .37 | .35 | .33 | .31 | .30 | .29 | .28 | .27 | .26 | .25 | .24 | .24 | .23 |
| 12 | 1.16 | .88 | .71 | .60 | .52 | .46 | .41 | .38 | .35 | .32 | .30 | .28 | .26 | .25 | .23 | .22 | .21 | .20 | .19 |
| | .88 | .63 | .54 | .47 | .43 | .39 | .37 | .35 | .33 | .31 | .30 | .28 | .27 | .26 | .25 | .25 | .24 | .23 | .23 |
| 13 | 1.17 | .89 | .72 | .61 | .53 | .47 | .42 | .38 | .35 | .32 | .30 | .28 | .26 | .25 | .24 | .22 | .21 | .20 | .19 |
| | .86 | .62 | .53 | .47 | .42 | .39 | .36 | .34 | .32 | .31 | .29 | .28 | .27 | .26 | .25 | .24 | .24 | .23 | .22 |
| 14 | 1.18 | .90 | .73 | .62 | .54 | .48 | .43 | .39 | .36 | .33 | .31 | .29 | .27 | .25 | .24 | .23 | .22 | .21 | .20 |
| | .85 | .62 | .52 | .46 | .42 | .39 | .36 | .34 | .32 | .30 | .29 | .28 | .27 | .26 | .25 | .24 | .23 | .23 | .22 |
| 15 | 1.19 | .91 | .74 | .63 | .55 | .48 | .43 | .39 | .36 | .33 | .31 | .29 | .27 | .26 | .24 | .23 | .22 | .21 | .20 |
| | .84 | .61 | .52 | .46 | .41 | .38 | .36 | .33 | .32 | .30 | .29 | .28 | .27 | .26 | .25 | .24 | .23 | .23 | .22 |
| 16 | 1.21 | .92 | .75 | .64 | .55 | .49 | .44 | .40 | .37 | .34 | .31 | .29 | .28 | .26 | .25 | .23 | .22 | .21 | .20 |
| | .82 | .60 | .51 | .45 | .41 | .38 | .35 | .33 | .31 | .30 | .29 | .27 | .26 | .25 | .25 | .24 | .23 | .23 | .22 |
| 17 | 1.22 | .93 | .76 | .65 | .56 | .50 | .44 | .40 | .37 | .34 | .32 | .30 | .28 | .26 | .25 | .24 | .23 | .22 | .21 |
| | .81 | .60 | .51 | .45 | .41 | .38 | .35 | .33 | .31 | .30 | .28 | .27 | .26 | .25 | .24 | .24 | .23 | .22 | .22 |
| 18 | 1.23 | .94 | .77 | .65 | .57 | .50 | .45 | .31 | .38 | .35 | .32 | .30 | .29 | .27 | .25 | .24 | .23 | .22 | .21 |
| | .81 | .59 | .50 | .45 | .40 | .37 | .35 | .33 | .31 | .29 | .28 | .27 | .26 | .25 | .24 | .24 | .23 | .22 | .22 |
| 19 | 1.24 | .95 | .78 | .66 | .57 | .51 | .46 | .42 | .38 | .35 | .33 | .31 | .29 | .27 | .26 | .24 | .23 | .22 | .21 |
| | .80 | .59 | .50 | .44 | .40 | .37 | .35 | .33 | .31 | .29 | .28 | .27 | .26 | .25 | .24 | .23 | .23 | .22 | .22 |
| 20 | 1.26 | .97 | .79 | .67 | .58 | .51 | .46 | .42 | .39 | .36 | .33 | .31 | .29 | .27 | .26 | .25 | .23 | .22 | .21 |
| | .79 | .58 | .50 | .44 | .40 | .37 | .34 | .32 | .31 | .29 | .28 | .27 | .26 | .25 | .24 | .23 | .23 | .22 | .21 |

The derivation and computation of TABLES 2 and 3 will be discussed in a forthcoming paper by Kurtz, Link, Tukey, and Wallace[3] and are discussed and compared with definitive and other competitive methods in an account entitled "The Problem of Multiple Comparisons" being prepared by the writer.[6]

*Further Examples*

We now treat a number of examples of increasing complexity quite briefly, illustrating the applicability of similar methods. The methods presented are chosen for (1) simplicity of use, and (2) transparency of calculation as to function and purpose. They are recommended "as is" to the

occasional analyst, as the writer's choice of today among available "quick and easy" methods. The professional analyst may prefer various modifications for good reasons.

All of the methods presented are "quick and easy," but to each there corresponds a definitive method. Many of these definitive methods are not at present in print, but will appear in the account mentioned above.[6]

*Example 2. Unbalanced single classification.* Tippett[5] gives 9 warp breakage rates for warps made with 6 yarns.

Let us suppose that 5 of these values were lost, strayed or stolen, so that the observations and analysis were as follows:

| | *AL* | *AM* | *AH* | *BL* | *BM* | *BH* | |
|---|---|---|---|---|---|---|---|
| | 26 | 18 | 36 | 27 | 42 | 20 | |
| | 30 | 21 | 21 | 14 | 26 | 21 | |
| | 54 | 29 | 24 | 29 | 19 | x | |
| | 25 | x | 18 | 19 | 16 | 17 | |
| | 70 | 12 | 10 | x | 39 | 13 | |
| | 52 | 18 | 43 | x | 28 | 15 | |
| | 51 | 35 | x | 41 | 21 | 15 | |
| | 26 | 30 | 15 | 20 | 39 | 16 | |
| | 67 | 36 | 26 | 44 | 29 | 28 | |
| Ranges of first 7 present | 45 | 23 | 33 | 30 | 26 | 8 | TR= 165 |
| Totals | 401 | 199 | 193 | 194 | 259 | 145 | |
| Cases | 9 | 8 | 8 | 7 | 9 | 8 | |
| Means | 44.6 | 24.9 | 24.1 | 27.7 | 28.8 | 18.1 | |

Here we have taken ranges of 7, since BL had only 7 observations, and have used the first seven observations available, thus assuming that the order in the column was either random, or the order in which the tests were made.

If we had had only 7 observations in each column, then the appropriate allowances would have been

for totals of 7: $165\,(0.69) = 112.7$,

for means of 7: $\frac{112.7}{7} = 16.1$.

For the actual means (involving 7 or more values) we are entitled to a slight reduction in the allowances (actually from 16.1 to 15.9) but if quick and easy analyses are in order, it is usually not worth while to calculate this reduction. (The argument, previously suggested by the writer, that 49 observations in place of 42 deserve an allowance $\sqrt{42/49}$ as long, which would lead to 14.9 in this example, turns out to be radical and dangerous.) If we do not try to apply this reduction, our error rates will be slightly conservative. Thus we conclude that

(1) the average difference between AL and BH is

$$44.6 - 18.1 \pm 16.1 = 26.5 \pm 16.1$$

$$= \text{between } +10.4 \text{ and } +42.6,$$

(2) the average difference between AM and AH is

$$24.9 - 24.1 \pm 16.1 = 0.8 \pm 16.1$$

$$= \text{between } -15.3 \text{ and } +16.9,$$

and so on.

*Example 3. Balanced double classification.* Goulden[2] gives duplicate loaf volumes, in coded form, for 5 varieties of wheat each grown at four stations. In this example we shall analyze, the (coded) total volumes (1 unit = 10 $cm^3$) for the pairs of loaves.

The analysis proceeds as follows:

| | (IV) | | I | | II | | III | | IV | | | Total |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | *(37.6)* | *-25.6* | 12.0 | *17.5* | 29.5 | *1.5* | 31.0 | *6.6* | 37.6 | | | 110.1 |
| 2 | *(48.2)* | *-22.5* | 25.7 | *12.8* | 38.5 | *-9.5* | 29.0 | *19.2* | 48.2 | | | 141.4 |
| 3 | *(36.3)* | *-28.3* | 8.0 | *10.0* | 18.0 | *11.5* | 29.5 | *6.8* | 36.3 | | | 91.8 |
| 4 | *(31.4)* | *-27.9* | 3.5 | *24.5* | 28.0 | *-10.5* | 17.5 | *13.9* | 31.4 | | | 80.4 |
| 5 | *(46.8)* | *10.2* | 57.0 | *-21.5* | 35.5 | *13.0* | 22.5 | *24.3* | 46.8 | | | 161.8 |
| | | *38.5* | | *56.0* | | *24.5* | | *17.7* | | *TR136.7* | .69 | *±94* |
| | | | | | | | | | | .72 | | |
| | | | 106.2 | | 149.2 | | 129.5 | | 200.3 | *±98* | | 585.5 |

Most of the italic numbers represent horizontal differences, the exception being the first column, which duplicates column IV. TABLE 3 supplies the factors 0.69 and 0.72 which are multiplied into the total range, 136.7, to obtain the WSD's ±94 and ±98.

We conclude, for example, that

(1) on the average, variety 1 exceeds variety 2 by

$$110.1 - 141.4 \pm 94 = \text{between } -125 \text{ and } +63 \text{ units}$$

in the volume of 8 loaves (between −156 and +79 $cm^3$ per loaf),

(2) on the average, location II exceeds location III by

$$149.5 - 129.5 \pm 98 = \text{between } -78 \text{ and } +110 \text{ units}$$

in the volume of 10 loaves (between −78 and +118 $cm^3$ per load),

(3) on the average, all varieties may yield the same loaf volume, in any event no two differ by more than

$$161.8 - 80.4 + 94 = 175 \text{ units}$$

in the volume of 8 loaves (by more than 218 $cm^3$ per loaf).

Here "on the average" has a very specific meaning. When the statement is about varieties, it means "averages over many loaves and over a large population of locations of which locations I, II, III, IV are a random sample." When the statement is about locations, it means "averaged over many loaves and over a population of varieties, of which varieties 1, 2, 3, 4, 5 are a random sample." Note that the averaging is over the other classification, and over a population of varieties or locations of which only a sample was measured.

By using TABLE 3, we spend a 5 per cent error rate on statements about varieties and another 5 per cent on statements about locations.

*Example 4. Slight irregularities in a double classification.* Let us suppose that the value of 57.0 for variety 5 at location I were missing. How then could we conduct the analysis?

If we drop all volumes for variety 5, we could find ranges of 5.8, 14.5, 27.0, and 12.6 for a total range of 54.9. TABLE 3 would supply factors of .76 and .76, yielding WSD's of ±47.2 for each set of totals (4 varieties at 4 locations).

If we drop all volumes for location I, we would find ranges of 10.2, 24.5, and 17.7 for a total range of 52.4. TABLE 3 would supply factors of .90 and .84 yielding WSD's of ±47.2 for location totals and ±44.0 for variety totals (5 varieties at 3 locations).

We may assemble all the relevant numbers in the following form:

TABLE FOR EXAMPLE 4

| Locations used: | I, II, III, IV | II, III, IV | all |
|---|---|---|---|
| Varieties used: | 1, 2, 3, 4 | 1, 2, 3, 4, 5 | all |
| Location means: | | | |
| I | 12.3 | x | 13.0 |
| II | 28.5 | 29.9 | 29.9 |
| III | 26.8 | 25.9 | 25.9 |
| IV | 38.4 | 40.1 | 40.1 |
| WSD | ±11.8 | x | ±11.8 |
| Variety means | | | |
| 1 | 27.5 | 32.7 | 27.5 |
| 2 | 35.4 | 38.6 | 35.4 |
| 3 | 23.0 | 27.9 | 23.0 |
| 4 | 20.1 | 25.6 | 20.1 |
| 5 | x | 42.1 | 37.4 |
| WSD | x | ±14.7 | ±14.7 |

Only two aspects of this table need particular explanation: (1) the means supplied for the variety and location encompassing the missing values and (2) the final WSD's. We have

$$\begin{aligned} 29.9 - 28.5 &= \phantom{-}1.4, \\ 25.9 - 26.8 &= -0.9, \\ 40.1 - 38.4 &= \phantom{-}1.7, \end{aligned}$$

so that the average change from a location mean over varieties 1, 2, 3, 4 to a location mean over varieties 1, 2, 3, 4, *and* 5 is

$$\frac{1.4 - 0.9 + 1.7}{3} = 0.7.$$

Thus

$$13.0 = 12.3 + 0.7$$

the adjustment from 42.1 to 37.4 is obtained similarly from

$$\frac{5.2 + 3.2 + 4.9 + 5.5}{4} = 4.7.$$

Next, observe that although ±11.8 is based on 16 of the 19 observations and although ±14.7 is based on only 15 of the 19 observations, we have not attempted to calculate and apply a corresponding reduction in the allowance. The analysis indicated here is only reasonable for one, or a very few missing values. Otherwise a more sophisticated analysis is likely to be needed.

*Example 5. Double classification with replication.* We now revert to the whole of the Goulden data, bringing forward the individual loaf volumes (still coded). The analysis runs as follows:

| *Loc.* | *Var.* | *Volumes* | | *Range* | *Total* |
|---|---|---|---|---|---|
| I | 1 | 7.5 | 4.5 | 3.0 | 12.0 |
| | 2 | 12.5 | 13.2 | 0.7 | 25.7 |
| | 3 | 7.0 | 1.0 | 6.0 | 8.0 |
| | 4 | 1.5 | 2.0 | 0.5 | 3.5 |
| | 5 | 28.0 | 29.0 | 1.0 | 57.0 |
| II | 1 | 15.5 | 14.0 | 1.5 | 29.5 |
| | 2 | 20.0 | 18.5 | 1.5 | 38.5 |
| | 3 | 10.0 | 8.0 | 2.0 | 18.0 |
| | 4 | 13.0 | 15.0 | 2.0 | 28.0 |
| | 5 | 17.5 | 16.0 | 3.5 | 35.5 |
| III | 1 | 16.5 | 14.5 | 2.0 | 31.0 |
| | 2 | 15.0 | 14.0 | 1.0 | 29.0 |
| | 3 | 15.5 | 14.0 | 1.5 | 29.0 |
| | 4 | 8.5 | 9.0 | 0.5 | 17.5 |
| | 5 | 10.5 | 12.0 | 1.5 | 22.5 |
| IV | 1 | 19.0 | 18.6 | 0.4 | 37.6 |
| | 2 | 23.8 | 24.4 | 0.6 | 48.2 |
| | 3 | 17.8 | 18.5 | 0.7 | 36.3 |
| | 4 | 14.8 | 16.6 | 1.8 | 31.4 |
| | 5 | 22.0 | 24.8 | 2.8 | 46.6 |

TR = 34.5
Factor = 0.36
WSD = ±12.4

We may set forth the results as follows:

| | | *I* | *II* | *III* | *IV* | *Means* | |
|---|---|---|---|---|---|---|---|
| | *1* | 12.0 | 29.5 | 31.0 | 37.6 | 27.5 | |
| | *2* | 25.7 | 38.5 | 29.0 | 48.2 | 35.4 | |
| (1) | *3* | 8.0 | 18.0 | 29.5 | 36.3 | 22.9 | ±12.4 |
| | *4* | 3.5 | 28.0 | 17.5 | 31.4 | 20.1 | pairwise |
| | *5* | 57.0 | 35.5 | 22.5 | 46.8 | 40.4 | |
| | *Means* | 21.2 | 29.9 | 25.8 | 40.1 | 29.3 | |

and conclude for example, that:

(1) on the average, 4II produces fluffier loaves than 1I by 28.0 – 12.0 ± 12.4 = 16.0 ± 12.4 = between 3.6 and 28.4 for the volume of two loaves.

(2) on the average, location IV produces fluffier loaves than location I by 40.1 – 21.2 ± 12.3 = 18.9 ± 12.4 = between +6.5 and +31.3 for the volume of two loaves.

(3) on the average variety 4 produces fluffier loaves than variety 2 by 20.1 – 35.4 ± 12.3 = –15.3 ± 12.4 = between –27.7 and –2.9 for the volume of two loaves (that is, variety 2 makes them fluffier on the average than variety 4).

In (2) and (3), "on the average" has very definite meanings, which are *very* different from those of example 3. In (2), and in other such statements about locations, "on the average" means "on the average over many loaves and these particular five varieties, each variety weighted equally." In (3), and in other such statements about varieties, "on the average" means "on the average over many loaves and these particular four locations, each location weighted equally." Note that the averaging is still over the *other* classification, but is now only over the *sample* of varieties or locations *actually* measured.

Statements like (2) and (3) are statements about *particular* averages. It is not surprising that we can say more about particular averages than about general averages.

*Example 6. Analysis of interactions.* We now continue with example 5, seeking manifest irregularities in the totals. If we subtract location means, we have

| | | *I* | *II* | *III* | *IV* | *Means* | (=variety mean deviations) |
|---|---|---|---|---|---|---|---|
| | *1* | -9.2 | -0.4 | 5.2 | -2.5 | -1.8 | |
| | *2* | 4.5 | 8.6 | 3.2 | 8.1 | 6.1 | |
| (2) | *3* | -13.2 | -11.9 | 3.7 | -3.8 | -6.4 | ±9.9 |
| | *4* | -17.7 | -1.9 | -8.3 | -8.7 | -9.2 | |
| | *5* | 35.8 | 5.6 | -3.3 | 6.7 | 11.1 | |

where ±9.9 = (1 – 1/5) (±12.4) and where the factor of 1 – 1/5 arises because the contrasts occurring have positive (and negative) coefficients adding up to 1 – 1/5. (In general, we may apply a factor of 1 – 1/(number per mean) when we take deviations from means.) Continuing by subtracting variety mean deviations, we find

| | | | | | | |
|---|---|---|---|---|---|---|
| | 1 | -7.4 | 1.4 | 7.0 | -0.7 | |
| | 2 | -1.6 | 2.5 | -2.9 | 2.0 | |
| (3) | 3 | -6.8 | -5.5 | 10.1 | 2.6 | ±14.9 |
| | 4 | -8.5 | 7.3 | 0.9 | 0.5 | |
| | 5 | 24.7 | -5.5 | -14.4 | -4.4 | |

where $\pm 14.9 = 2(1 - 1/4)(\pm 9.9)$. (The additional factor of 2 is required each time deviations are taken from *deviations*.) Thus we see that I5 yields unexpectedly heavy loaves by an amount $24.7 \pm 14.9$ (between 9.8 and 39.6).

We could have reached the same values by first subtracting variety means to obtain

| | | *I* | *II* | *III* | *IV* | |
|---|---|---|---|---|---|---|
| | *1* | -15.5 | 2.0 | 3.5 | 10.1 | |
| | *2* | -9.7 | 3.1 | -6.4 | 12.8 | |
| (4) | *3* | -14.9 | -4.9 | 6.6 | 11.4 | ±9.9 |
| | *4* | -16.5 | 7.9 | -2.6 | 11.4 | |
| | *5* | 17.0 | -4.9 | -17.9 | 6.4 | |
| | *Means* | -7.9 | 0.6 | -3.4 | 10.8 | |

and then subtracting location mean deviations to obtain

| | | *I* | *II* | *III* | *IV* | |
|---|---|---|---|---|---|---|
| | *1* | -7.6 | 1.4 | 6.9 | -0.7 | |
| | *2* | -1.8 | 2.5 | -3.0 | 2.0 | |
| (5) | *3* | -7.0 | -5.5 | 10.0 | 2.6 | ± 14.9 |
| | *4* | -8.6 | 7.3 | 0.8 | 0.6 | |
| | *5* | 24.9 | -5.5 | -14.5 | -4.4 | |

the small difference between array (3) and array (5) are due to rounding (we carried the means and mean deviations to only one decimal place) and are of no practical significance. (Making the computation both ways affords an excellent check.)

If we admit that I5 is definitely out of line, we may well wish to reanalyze the observations leaving it aside. The values in columns II, III, IV of (2) are unaffected by I5, so we may find

| | | *II* | *III* | *IV* | *Means* | |
|---|---|---|---|---|---|---|
| | *1* | -0.4 | 5.2 | -2.5 | 0.8 | |
| | *2* | 8.6 | 3.2 | 8.1 | 6.6 | |
| (6) | *3* | -11.9 | 3.7 | -3.8 | -4.0 | ±9.9 |
| | *4* | -1.9 | -8.3 | -8.7 | -6.3 | |
| | *5* | 5.6 | -3.5 | 6.7 | 3.0 | |

and continue by subtracting variety mean deviations to find

| | | *II* | *III* | *IV* | |
|---|---|---|---|---|---|
| | *1* | -1.2 | 4.4 | -3.3 | |
| | *2* | 2.0 | -3.4 | 1.5 | |
| (7) | *3* | -7.9 | 7.7 | 0.2 | ±14.9 |
| | *4* | 4.4 | -2.0 | -2.4 | |
| | *5* | 2.6 | -6.3 | 3.7 | |

Similarly, the values in rows 1, 2, 3, 4 of (4) are unaffected by I5, so we may find

| | | *I* | *II* | *III* | *IV* | |
|---|---|---|---|---|---|---|
| | *1* | -15.5 | 2.0 | 3.5 | 10.1 | |
| | *2* | -9.7 | 3.1 | -6.4 | 12.8 | |
| (8) | *3* | -14.9 | -4.9 | 6.6 | 13.4 | ±9.9 |
| | *4* | -16.5 | 7.9 | -2.6 | 11.4 | |
| | *Means* | -14.1 | 2.0 | 0.3 | 11.9 | |

and continue by subtracting location mean deviations to find

| | | *I* | *II* | *III* | *IV* | |
|---|---|---|---|---|---|---|
| | *1* | -1.4 | 0.0 | 3.2 | -1.8 | |
| | *2* | 4.4 | 1.1 | -6.7 | 0.9 | |
| (9) | *3* | -0.9 | -6.9 | 6.3 | 1.5 | ±14.9 |
| | *4* | -2.4 | 5.9 | -2.9 | -0.5 | |

Now if we take the mean of (7) and (9) where they overlap, and the individual values where they do not, we find

| | *I* | *II* | *III* | *IV* | |
|---|---|---|---|---|---|
| *1* | -1.4 | -0.6 | 3.8 | -2.5 | |
| *2* | 4.4 | 1.6 | -5.0 | 1.2 | |
| *3* | -0.9 | -7.4 | 7.0 | 0.8 | ±14.9 |
| *4* | -2.4 | 5.2 | -2.4 | -1.4 | |
| *5* | x | 2.6 | -3.4 | 3.7 | |

which is to be reasonably interpreted as indicating that, except for I5, interactions *may* be absent from this situation, and that it is fairly sure that they are less than $7.4 + 14.9 = 22.3$ in absolute value.

More refined methods are available for use when necessary.

*Example 7. Gaps.* It is sometimes desired to make separations into groups rather than to set limits on differences. This may be done by identifying gaps. The required length of apparent gaps depends on the WSD as follows:

| *Number compared* | *Gap required* | *Number compared* | *Gap required* |
|---|---|---|---|
| 1 | | 11 | .80 WSD |
| 2 | 1.00 WSD | 12 | .80 WSD |
| 3 | 0.83 WSD | 13 | .79 WSD |
| 4 | .86 WSD | 14 | .80 WSD |
| 5 | .81 WSD | 15 | .79 WSD |
| 6 | .84 WSD | 16 | .80 WSD |
| 7 | .81 WSD | 17 | .79 WSD |
| 8 | .82 WSD | 18 | .79 WSD |
| 9 | .80 WSD | 19 | .78 WSD |
| 10 | .81 WSD | 20 | .79 WSD |

If gaps above are used, the error rate for

gaps alone will be 5 per cent in the most unfavorable circumstances.

If gaps are combined with allowances, the over-all error rate will be about

| | | | |
|---|---|---|---|
| 5.5% | for | 2 | totals or means |
| 7.5% | for | 8 | totals or means |
| 10. % | for | many | totals or means |

in the most unfavorable situation.

Returning to the first example, the means, WSD and significant gap are

| | | |
|---|---|---|
| Sample | 1 | 1505 |
| | 2 | 1528 |
| | 3 | 1564 |
| | 4 | 1498 |
| | 5 | 1600 |
| | 6 | 1470 |
| WSD | | ±91 |

Required gap = (0.84) (91) = 76

Using gaps above we draw no definite conclusions, and are entitled to make no further statements. (Using gaps and allowances we come to the same conclusions as with allowances alone.)

*References*


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## *SECTION OF ANTHROPOLOGY**

### HUMAN COLOR PERCEPTION AND BEHAVIORAL RESPONSE †

*By* VERNE F. RAY ‡

In his introductory remarks to a recent symposium on perception Heinz Werner pointed out that the man-in-the-street is quite sure that he sees with his eyes and hears with his ears.[1] Apparently this naive view is shared by anthropologists. It is generally futile to search in an ethnography for so much as a paragraph, to say nothing of a chapter, on the subject of perceptual patterning as affected by the particular culture under discussion. Theoretical or analytical papers on the subject are equally rare. A good start was made by W. H. R. Rivers over forty years ago but his stimulating leads were never followed up by his contemporaries or successors.

Psychologists, on the other hand, have always given generous attention to problems of perception. Currently these studies are concerned largely with the projective nature of perception. An attempt is being made to determine the manner in which "the perceived world pattern mirrors the organized need pattern within," as Gardner Murphy has phrased it.[2]

These studies should be of great interest to the anthropologist; they naturally focus upon the individual but the basic problem is one which demands an analysis of the cultural factors operating before it can be answered. The organized need pattern within the person is based largely upon the organized pattern of his culture, only secondarily upon individually unique learning. Psychologists must usually, if not always, study narrow individual directive states which are mere variations upon a theme. The theme itself and all comparable themes are cultural in character and the notes constituting them, indeed the character of the bars upon which they are written, can only be described and interpreted by anthropologists. In neglecting our duty we not only deprive ourselves of significant insights into man's behavior but we put psychologists at an unfortunate disadvantage, that of working without adequate controls.

Let us take an example from a recent psychological experiment by Bruner and Postman.[3] Subjects at Radcliffe and Harvard were shown, successively, by tachistoscopic exposure, five different playing cards. On one to four of the cards color and suit were reversed; the normal and distorted cards were presented in random order. One interesting result was disruption, that is, gross failure to organize the perceptual field, with consequent frustration and inability to describe that which had been presented. When the subject was able to recognize a normal card in 20 to 50 milliseconds the same subject might despair of describing a distorted card after a hundred or more milliseconds of exposure. Such a state of frustration is brought about by the subject's failure to confirm any of his repertory of experiences. As a consequence, perception fails to produce any kind of meaningful behavior. "Repertory of experiences" and "meaningful behavior" are significant terms here. What repertory of experiences? That which derives from the subject's unique psychological makeup? No; it is rather the repertory of his culture that is the critical factor in his reaction. His conditioning in this respect—and his directive state—are derived from his culture and are shared by his fellows regardless of individual variations in experience.

* The Division of Mycology held a meeting on November 27, 1953 at which a paper, entitled "Dermatophytes in Lesions and in Cultures," was read by Emanuel Muskatblit. No abstract of this paper had been received at the time of publication.

† This paper was presented at a meeting of the Section on November 23, 1953.

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Meaningful behavior? Here again we are dealing with a notion which has significance at the cultural level. The playing card is a good example of a culturally arbitrary and symbolic object of perception. A person whose culture lacks playing cards would not be perturbed; for him there would be no perceptual disruption because no prescribed and therefore meaningful behavior is elicited by the perception of a black spade or a red diamond.

And thus is pointed the way to anthropologists for significant investigations of perceptual problems, investigations broad in base and experimental in method.

These introductory remarks have had to do with vision simply because the great bulk of work on perception has been concerned with seeing in its various manifestations. The complexity of this phase of perception and the intriguing problems it presents make this predilection understandable but hardly defensible because we are left with a woefully inadequate knowledge of perception via the other senses.

My own experimental and field researches have included studies of auditory perception but the bulk of my work has been in the field of vision. I have attempted to develop an adequate methodology and technique for the anthropological study of human color vision and to advance somewhat our knowledge of that aspect of perception. Toward these ends I have worked from time to time over the last twenty years with subjects, mostly American Indians, representing about sixty linguistically and culturally distinct groups. The diversity is indicated by the fact that approximately twenty linguistic stocks were involved, in terms of the old Powell classification, and six culture areas have been covered. In a recent paper I summarized my recommendations regarding technique and gave some attention to methodology and problems.[4] At this time I wish to present some of my experimental data but first it may be wise to repeat a word or two about technique.

After experimenting with spectroscopes and spectrographs and finding them unsatisfactory I sought other means for obtaining perceptual data. I demanded a technique which would permit designation of color quality in terms of wavelength since only in this way would it be possible to convey the results of my findings to fellow workers. Attempts to establish international color standards have failed completely and the reproduction of color samples to accompany a scientific article would be either technically impossible or prohibitively expensive. The most desirable color sample for perceptual research is a pigmented flat surface. Contrary to expectation, it was found possible to produce such samples with a purity and uniformity more than adequate for use in perceptual experiments and at the same time identifiable in terms of wavelength. In experimentation these samples are presented to subjects under rigidly controlled conditions as to quality of light, background, method of exposure, character of responses, recordings of responses—and, of course, the samples are presented in random order. The basic response of the subject is the identification of the color perceived by the appropriate color term in his particular language. In so far as the scientific analysis of the data is concerned color names are dispensed with entirely and wavelength designations used exclusively. However, for purposes of writing and discussion, it is desirable to have a name designation to correspond with each of the wavelength numbers. Standard color names can be used with the understanding that they mean nothing more than a wavelength span. A particular wavelength and name designates a specific point in a continuum, the spectrum. It would be preferable to have the dividing lines between one sample and the two adjacent samples identified but this is impossible with discrete samples, possible only with an elaborate spectroscope or a continuous spectral reproduction; the former is impra-

ticable, the latter is useless for these experimental purposes because of the carry-over or contamination effect of the visible adjacent colors. However, the various samples are arbitrary as to wavelength position, and the number of samples is generally much greater than the basic color-name repertory of the culture. Therefore an approximation of the wavelength position of the dividing lines can usually be made for any particular system. This is significant because the range for a particular cultural designation is sometimes narrow, sometimes broad, and the type or focal point is not always the mid-point—indeed it is often found at one or another extreme.

The accompanying chart (FIGURE 1) should of course be drawn as a circle or a wheel rather than linearly. The spectral continuum turns back upon itself, so to speak, in all systems in which the classification is extended to include the non-spectral violets, thus joining the two "ends" of the distribution. A circular drawing would avoid the awkwardness involved in repeating one of the violets at the two "ends" of a straight-line scale. But I have found that circular charts are hard to read and to conceptualize and for these reasons the linear presentation is to be preferred.

The position of the color name on the scale indicates the recognized type position for that particular color. The horizontal lines on either side of the color name indicate the range. A color name will sometimes be found opposite the dividing line between two of the segments into which the spectrum has been divided for experimental purposes. This means that the color samples on either side of the line are perceived as equally typical of the color in the culture concerned. It is not, of course, necessary to use color terms to indicate the structures of the various systems; a simple check mark or dot would serve the purpose. However, I have chosen to give the names for the interest that they may hold and for illustration of some factors which are linguistic in character.

The color systems presented are those of ten cultures of northwestern America: nine Indian and one American. The cultures vary in character from that of the northwestern United States to that of the Atka Eskimo and from Wishram Chinook to Chilcotin Athapaskan. The languages of the ten cultures are mutually unintelligible; several distinct stocks are represented. The cultures are not arranged in geographical order. Such an arrangement is less illuminating than one, as used here, which places together those systems which show the greatest similarity, especially as regards continuity of the horizontal lines which mark the boundaries of color range. The chart has been simplified to show only the standard colors, not the tints, shades or grays. If these were included the complexity of each system would be greatly increased.

This accounts only in part for the apparent simplicity of these systems. The chart shows only the basic color terms and the spectral bands which they designate. Hundreds or thousands of additional color descriptive terms are available to the users of each of these languages. But the significant point is that these are not color terms proper. They do not refer to recognized divisions of the color range conceived abstractly. They are terms such as mauve or chartreuse in English and each one has connotations beyond that of color. Sometimes the color meaning is dominant, other connotations secondary, as in the term scarlet; in other instances the reverse as, for example, smoky. In many cases the number of primary or secondary connotations is considerable; an example is chestnut. It is not uncommon to find true complexes of meaning, as witness sable. Some terms exhibit a fixed linkage of primary and secondary meanings; for instance, sorrel. In the vast majority of cases it is difficult or impossible to determine whether color connotation is primary or secondary; consider chartreuse, mauve, and cardinal, to name but a few.

These examples from English could be

duplicated in kind in any one of the languages represented on the chart. Naturally they are more numerous in some languages than others. The primary reason for the variation is cultural tolerance or encouragement, not linguistic structure. Some languages provide mechanisms for the invention of new color terms on the spur of the moment, for example blue-blue-violet. However, these linguistic devices relate to true color terms as opposed to the color descriptive words and phrases that we are now discussing.

The most significant characteristic of color descriptors – to coin a word – is the fact that they form no system; they are merely a random collection of useful words gathered from diverse sources. As such there is an amazing tolerance to their careless usage – one can hardly speak of inaccurate usage – and consequently great variation from person to person.

The perception of colors in descriptive terms is an exceedingly complex phenomenon. When I see a brightly-colored sunset my behavioral response, if linguistic, may not involve color at all but, instead, fire, waves, clouds, dusk, autumn, beauty, music, rest. Or a rose: beauty, odor, loveliness, softness, dew. And if color response be involved it is apt to be in terms of descriptors, not color terms. The latter relate to concepts, the former to things.

Perhaps the significance of what I have called color terms proper and the basic color system is now more clear. Perception of color and behavioral response thereto is our problem; not description of color. The basic color system is the foundation of precise behavior with respect to the world of color. The cultural repertory of experience provides appropriate behavior – utterance of the proper color term, for example – when perception occurs. No individual variation is permitted within the culture in this respect, except it be interpreted as error, and all normal persons perceive nearly equally well.

I have never encountered a culture lacking such a basic system. It is not an easy task to separate proper color terms from words which are merely descriptors, especially when the culture and its language are not too well known. I doubt that it can be done with an acceptable degree of validity without experimentation of the general type which I have described. Such a technique is effective in this respect because it brings out the system without the subject being aware that a system is being sought; he is asked no questions whatsoever. The procedure is quite analogous to that of the linguist who derives the grammar of a language from what the informant thought were random words, phrases and texts. In each case the problem of analysis is considerable and the end product often less than perfect. But the fundamental characteristics of the pattern are clearly exhibited in both instances.

Let us now return to the chart and see what variations in pattern are found. First we may look for instances of cross-cultural uniformity. None is found; the closest approach is in the region of red. Next we see that several cultures, the Tenino, Atka, Chetco, and Kalapuya, exhibit what has been called "blue-green confusion." Before discussing this let us look at the American perceptual pattern. We see that the green band is exceptionally narrow and that the type is immediately adjacent to blue. Only the Chilcotin draw the line where we do. The Tenino, for example, place the boundary just one unit from ours and thereby become guilty of "blue-green confusion" despite the fact that theirs is a more evenly divided system. But they are also victims of "green-yellow confusion" because they are not as "blue-green confused" as, say, the Atka who include all the greens with blue. The conclusion would seem to be that a color system without any "confusion" would have to possess a great number of basic terms – more than the number actually found in any recorded system – and the consequence of that might well be a hopeless individual confusion in

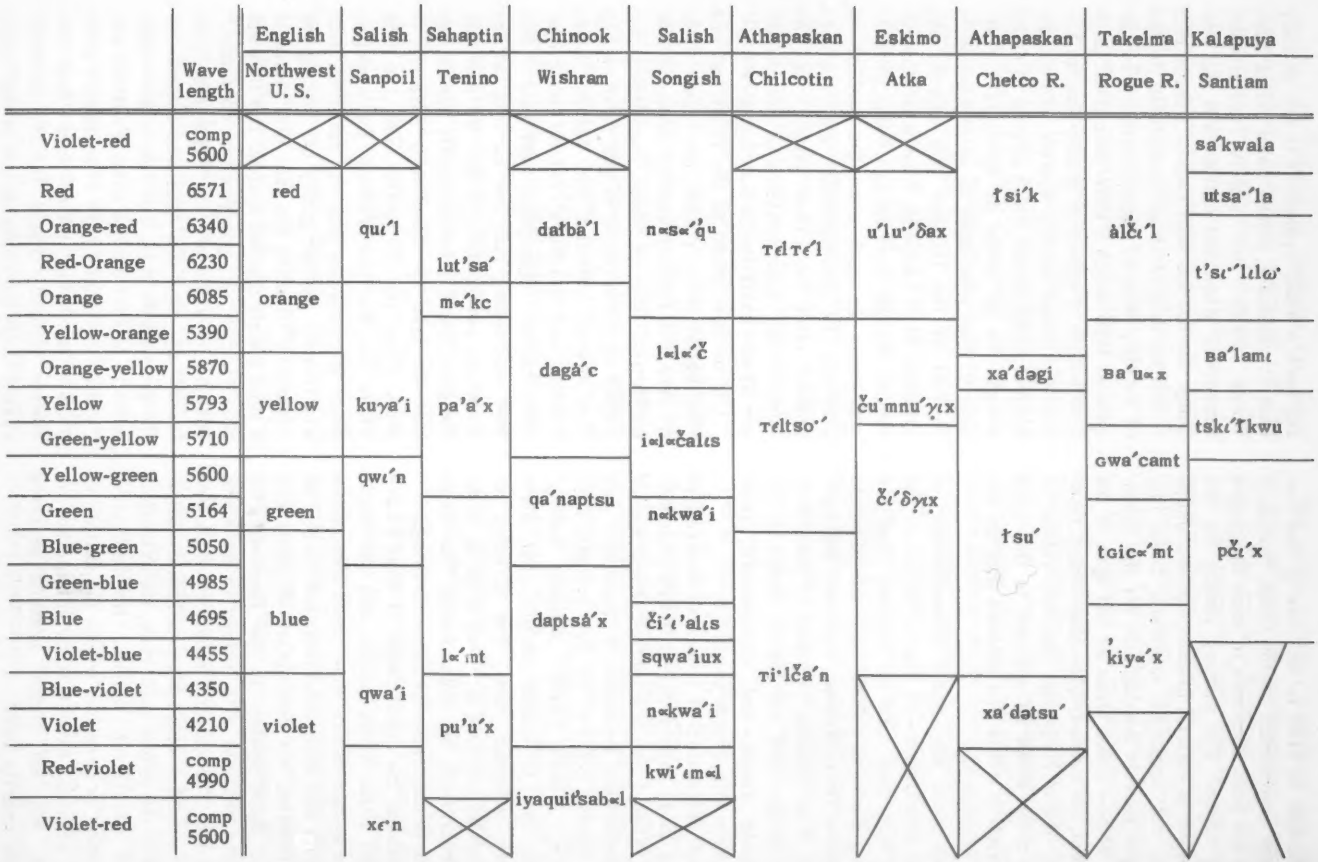
English
Northwest U. S.
Salish
Sanpoil
Sahaptin
Tenino
Chinook
Wishram
Salish
Songish
Athapaskan
Chilcotin
Eskimo
Atka
Athapaskan
Chetco R.
Takelma
Rogue R.
Kalapuya
Santiam
Wave length
Violet-red
comp 5600
Red
6571
Orange-red
6340
Red-Orange
6230
Orange
6085
Yellow-orange
5390
Orange-yellow
5870
Yellow
5793
Green-yellow
5710
Yellow-green
5600
Green
5164
Blue-green
5050
Green-blue
4985
Blue
4695
Violet-blue
4455
Blue-violet
4350
Violet
4210
Red-violet
comp 4990
Violet-red
comp 5600
red
orange
yellow
green
blue
violet
quɪ́ʼl
kuɣaʼi
qwɪ́ʼn
qwaʼi
xɛʼn
lutʼsaʼ
mɑ́ʼkc
paʼaʼx
lɑ́ʼɪnt
puʼuʼx
dałbàʼl
dagáʼc
qaʼnaptsu
daptsáʼx
iyaquitʼsabɑl
nɑsɑ́ʼq̓u
lɑlɑ́ʼč
iɑlɑčalɪs
nɑkwaʼi
čiʼɪʼalɪs
sqwaʼiux
nɑkwaʼi
kwiʼɛmɑl
ʇɛlʇɛʼl
ʇɛltsoʼʼ
ʇiʼlčaʼn
uʼluʼʼδax
čuʼmnuʼɣɪx
čɪʼδɣɪx
ʇsiʼk
xaʼdəgi
ʇsuʼ
xaʼdətsuʼ
àlčɪʼl
ʙaʼu=x
gwaʼcamt
tɑic=ʼmt
kiy=ʼx
saʼkwala
utsaʼʼla
tʼsɪʼʼlɪlɑwʼ
ʙaʼlamɪ
tskɪʼʇkwu
pčɪʼx

FIGURE 1.

perception and response. There is no question but that the Atka perceive broad bands of blue and green as one color just as the Sanpoil perceive orange and yellow to be unitary; the Chilcotin, blue and violet; the Chetco, red and orange; and the Americans ʁa'lamɪ and tskɪ'ɬkwu (Santiam). We designate the latter two colors by the unitary term yellow. This is the so-called "ʁa'lamɪ–tskɪ'ɬkwu confusion."

"Blue-green confusion" is often attributed to peoples who, as a matter of fact, perceive more subtly the variations in this region of the spectrum than we do. Instead of a dual division of the spread from the 5600 to 4455 wavelength positions, three divisions are perceived. When the naive investigator or observer notes that the middle segment of this tripartite division includes portions of what we call blue and green he tends to explain this phenomenon as confusion. A good example of the tripartite classification is found in the Songish system, a system which is characterized throughout by the subtlety of its distinctions. By what appears to be an accident of selection of examples, the chart shows only one other example of this pattern which is really quite common; this from the Rogue River.

Even the small amount of evidence presented on the chart demonstrates the low correlation of type and mid-point of range in color perception. In the American system the type for red is found at one end of the spectral range, green at the opposite end of its range. Tenino and Chetco present significant examples of types at one or another extreme of very broad bands. Chilcotin, on the other hand, is consistent in choosing the mid-point even though this culture is Athapaskan in speech as is Chetco. Frequently the type for a color in one system is located exactly where a division between colors is found in another pattern. Examples appearing on the chart are found in the Santiam column, also Wishram, Songish, and Tenino (5600-5164), Tenino and U. S. (4455-4350), Tenino and Sanpoil and others (4210) and in numerous other cases.

It is worthy of note that the number of terms in the basic systems shown varies only from three to eight with the mode at five and the average exactly five; the American pattern is close, with six. These figures would mean little, due to the small number of cases, were it not that they are consistent with most of the results shown by analysis of the total of sixty recorded cultures. These figures throw a new light upon the so-called primitive or simple systems which get along with three basic units. In the arbitrary division of the color continuum into three or eight segments there is a difference of finesse, not of kind, between the two, when it be a question of utility of the system. Behavior differs depending upon the perceptual grading: the person who has but three basic color terms at his disposal must more frequently use color descriptors to express his meaning. But behavior is meaningful in both cases; there is no disruption. Whether perception differs in the two instances is a matter which will be touched upon below.

Attempts to find correlations between the basic number of units in the color perception pattern and complexity of culture or development of art are doomed to failure if the sixty cultures studied are representative. It is true that the Chilcotin, with three basic terms, is at the same time one of the meagerest cultures represented on the chart but this cannot be said of the Atka. The most complex Indian culture presented is perhaps the Songish and it is also the one with the most numerous color distinctions. But American culture is far and away the most complex and artistically the most highly developed of these examples and we have but six categories to the Songish eight.

A search for environmental explanations is equally fruitless. The Atka have no basic color categories for the violets yet

it is precisely these colors which are predominant in their Aleutian environment. The environments of the Songish, Chetco, Rogue and Santiam are virtually identical but their color systems vary as widely as any recorded.

It will be seen that even a small quantity of data, as in this chart, provides numerous illustrations of fundamental aspects of color perception and response. With a few more researchers at work and a few more years of time we should have answers to numerous basic problems of culture and perception. And, in the course of such research, a great many related questions would be answered. Rough form can be given to some of these even now. Here are a few: Does human color perception have a peculiar linear and unidimensional quality, as contrasted to perception in terms of such multidimensional descriptors as those mentioned earlier? If so, is this quality characteristic also of other phenomena sharing this linear quality – in this instance, wavelength – for example, musical tones? Is man's richness of perception limited by behavioral response in terms of pattern? Is man's unique possession, language, a deterrent to full and varied response to his environment since its patterns often provide but few categories – as in color – whereas his physical sensitivity is exceedingly subtle? Does the artist see the world more richly and rewardingly because he divests himself of cultural pattern, and thus becomes less of a human being? Whatever the answer to these last questions may be, we must remember that if language makes possible human patterns, then pattern makes possible science; and also, that man is not a slave to pattern since he can perceive, and even respond in words, to the sunset or the rose as I have suggested is his wont.

Finally, I would like to repeat some of the conclusions which I offered in my earlier paper in advance of presentation of the experimental evidence. I conclude that there is no such thing as a "natural" division of the spectrum. The color systems of man are not based upon psychological, physiological, or anatomical factors. Each culture has taken the spectral continuum and has divided it upon a basis which is quite arbitrary except for pragmatic considerations. None of these systems derives from physiological limitations; none exploits more than meagerly the physiological sensitivity of the human being. The notion of color confusion reflects only the confusion of those expressing the idea.

Color systems serve to bring the world of color sensation into order so that perception may be relatively simple and behavioral response, particularly verbal response and communication, may be meaningful.

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## *CONFERENCE HELD*

### SECTION OF PHYSICS AND CHEMISTRY

"PROPERTIES OF SURFACES"

NOVEMBER 5 AND 6, 1953

The Section of Physics and Chemistry held a Conference on "Properties of Surfaces." Doctor Cecil V. King, New York University, New York, was the Conference Chairman in charge of the meeting.

The program consisted of the following papers:

*Thursday, November 5*

*Morning Session.*

"The Life History of Adsorbed Atoms, Ions, and Molecules," J. A. Becker, Bell Telephone Laboratories, Murray Hill, New Jersey.

"Interactions Between Solids and Gases and Surface Heterogeneity in Physical Adsorption," J. M. Honig, Purdue University, Lafayette, Indiana.

"Kinetics of Surface Processes," H. A. Taylor, New York University, New York, N.Y.

"The Effect of Monolayers on the Rate of Evaporation of Water," R. J. Archer and V. K. LaMer, Columbia University, New York, N.Y.

*Afternoon Session.*

"The Classical Theory of Diffusion as Applied to the Oxidation of Metals," E. A. Gulvransen, Westinghouse Research Laboratories, East Pittsburgh, Pennsylvania.

"Passivity in Metals and Adsorption," H. H. Uhlig, Massachusetts Institute of Technology, Cambridge, Massachusetts.

"Film Composition Studies," T. N. Rhodin, Jr., Du Pont Experimental Station, Wilmington, Delaware.

"Electron Microscope Surface Studies," C. J. Calbrick, Bell Telephone Laboratories, Murray Hill, New Jersey.

*Friday, November 6*

*Morning Session.*

"The Adsorption of Dyes to Silver Halide Microcrystals," W. West, Kodak Research Laboratories, Rochester, New York.

"Adsorption and Exchange in Metal-Metal Ion Systems," C. V. King, New York University, New York, New York.

"Adsorption and Decomposition of Vapors at a Germanium Surface," J. T. Law, Bell Telephone Laboratories, Murray Hill, New Jersey.

"Electrical Conductivity of Germanium Surfaces," E. N. Clarke, Sylvania Electric Products, Inc., Bayside, New York.

*Afternoon Session.*

"Exploration of Metal Surfaces with Fine Wires," R. H. Savage, General Electric Company, Schenectady, New York.

"Surface Properties of Silicon and Germanium," W. H. Brattain, Bell Telephone Laboratories, Murray Hill, New Jersey.

"Stabilization of Metallic Carbides by Non-Metallic Elements," H. H. Podgurski, Du Pont Experimental Station, Wilmington, Delaware.

The Section of Physics and Chemistry provides conferences for active workers in the special fields of Physics and Chemistry.

Attendance is limited to those invited to participate in these conferences and to interested Members of the Academy.

## *NEW MEMBERS*

*Elected November 19, 1953*

### LIFE MEMBERSHIP

Geschickter, Charles F., M.D., Medicine. Professor of Pathology, Georgetown University, Washington, D.C.

Jelinek, Bohdan, D.Sc., Biochemistry. Associated Professor of Biochemistry, University of Missouri, Columbia, Mo.

### SUSTAINING MEMBERSHIP

Halley, Henry, M.D., Pathology. St. Joseph Hospital, Alton, Ill.

Rozsa, George L., M.D., Morphology, Pathology. Associate Cancer Research Pathologist, Roswell Park Memorial Institute. Buffalo, N.Y.

### ACTIVE MEMBERSHIP

Allen, Paul, Jr., Chemistry. Ph.D., Professor of Chemistry, Stevens Institute of Technology, Hoboken, N.J.

Alperin, Irwin M., M.S., Marine Fisheries Biology. Aquatic Biologist, State of New York Conservation Department, Bureau of Marine Fisheries, Freeport, N.Y.

Alpher, Isadore M., M.D., Internal Medicine. Washington, D.C.

Barbar, Saul B., B.S., Sensory Physiology. Instructor in Biology, Williams College, Williamstown, Mass.

Beckett, George S., Civil Engineering. New York, N.Y.

Bergel, Meny, M.D., Cancer. Post-Doctoral Fellow, University of Rochester, Rochester, N.Y.

Bodemer, Charles W., M.A., Embryology, Anatomy. Graduate Assistant, Cornell University, Ithaca, N.Y.

Bonkalo, Alexander, M.D., Neuropsychiatry. Clinical Teacher, Research Assistant, University of Toronto, Toronto, Canada.

Brownell, Lloyd E., Ph.D., Sterilization with Gamma Radiation. Director of Fission Products Laboratory, University of Michigan, Ann Arbor, Mich.

Burch, Helen B., Ph.D., Nutritional Biochemistry. Research Associate, Washington University, St. Louis, Mo.

Clark, R. Lee, Jr., Surgery, Cancer. Director and Surgeon in Chief, University of Texas M.D. Anderson Hospital, Houston, Texas.

Cohn, Isidore, M.D., Surgery. Professor Clinical Surgery, Tulane University, New Orleans, La.

Cornwell, Charles D., Ph.D., Chemical Physics. Instructor, Physical Chemistry, University of Wisconsin, Madison, Wis.

Cox, Charles D., Ph.D., Medical Bacteriology. Professor and Chairman, Department of Microbiology and Public Health, University of South Dakota, Vermillion, S.D.

Cranford, Franzo Hazlett, Ph.D., Molecular Spectroscopy. Professor and Chairman, Department of Physics, Williams College, Williamstown, Mass.

Cuthbertson, Arnold Craig, Ph.D., Chemistry of Coal. Carnegie Professor of Chemistry, Mt. Allison University, Sackville, N.B., Canada.

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Fodor, Andrew, M.S., Microbiology. Chief of Clinical Pathology Division, First Army Area Medical Laboratory, New York, N.Y.

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Frommeyer, Walter Benedict, M.D., Internal Medicine, Hematology. Associate Professor of Medicine, Medical College of Alabama, Birmingham, Ala.

Gelfand, Maxwell L., M.D., Internal Medicine. Assistant Clinical Professor of Internal Medicine, New York University Post-Graduate Medical School, New York, N.Y.

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Department of Biochemistry, Margaret Hague Maternity Hospital, Jersey City, N.J.

Heinman, William Stettin, Engineering. New York, N.Y.

Hodgson, Roland, Ph.D., Biochemistry. Indianapolis, Ind.

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Juliet, Eduardo, M.D., Biology, Physics, Chemistry, Medical Director for Latin-America, Smith Kline and French Inc., Philadelphia, Pa.

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